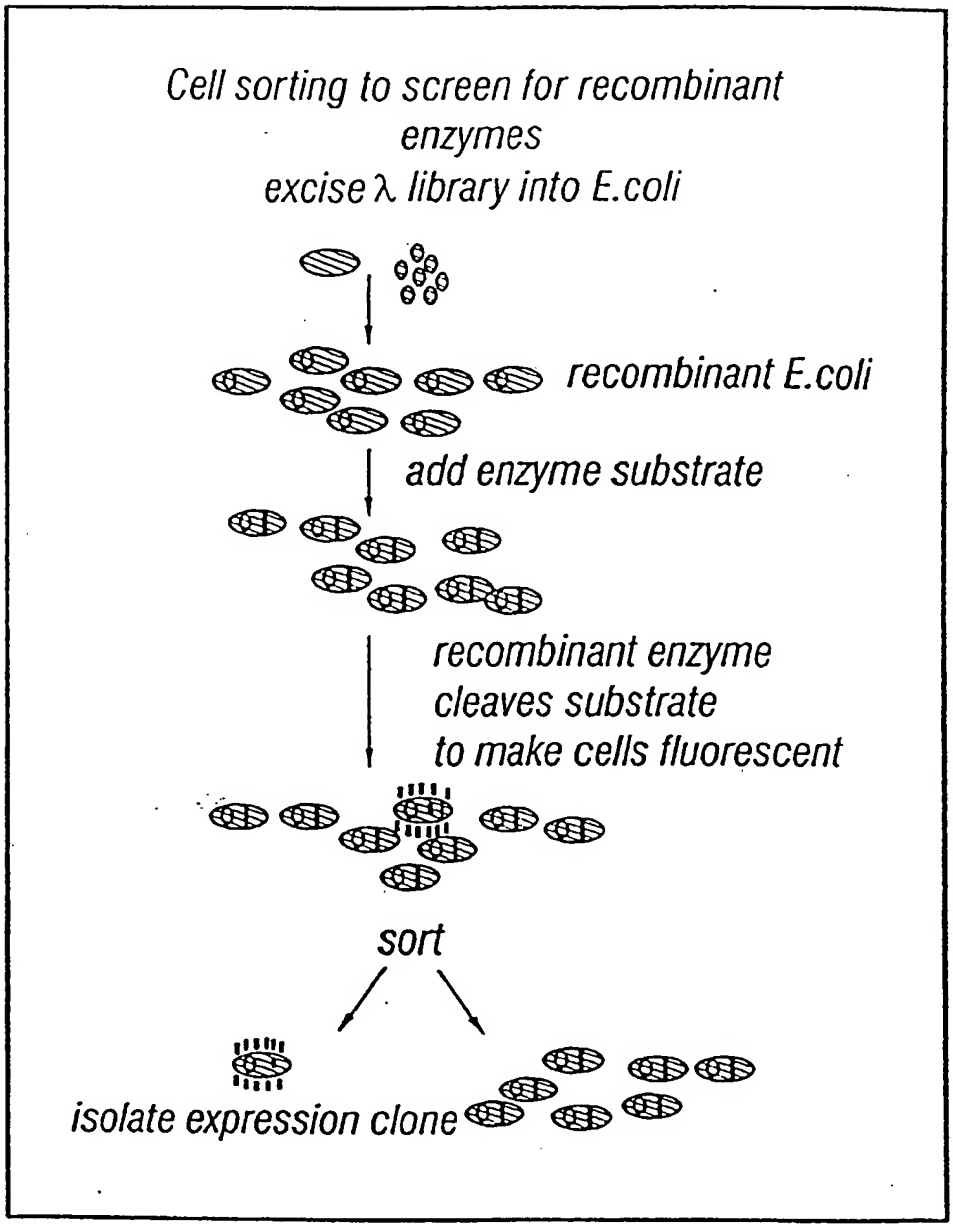

Cell sorting to screen for recombinant enzymes
excise λ library into E.coli
recombinant E.coli
add enzyme substrate
recombinant enzyme cleaves substrate to make cells fluorescent
sort
isolate expression clone

FIG. 1

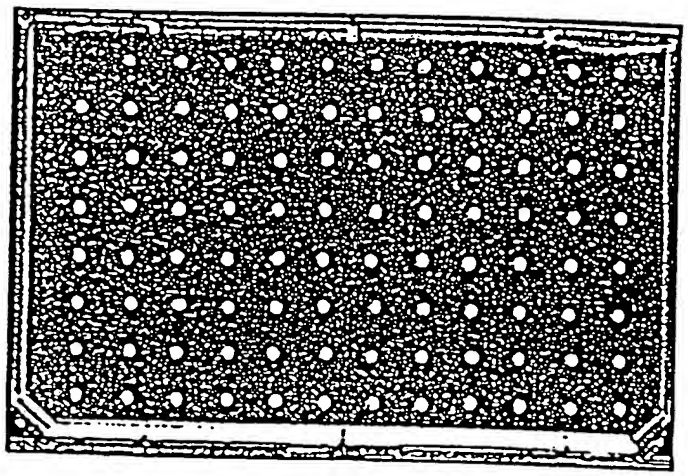
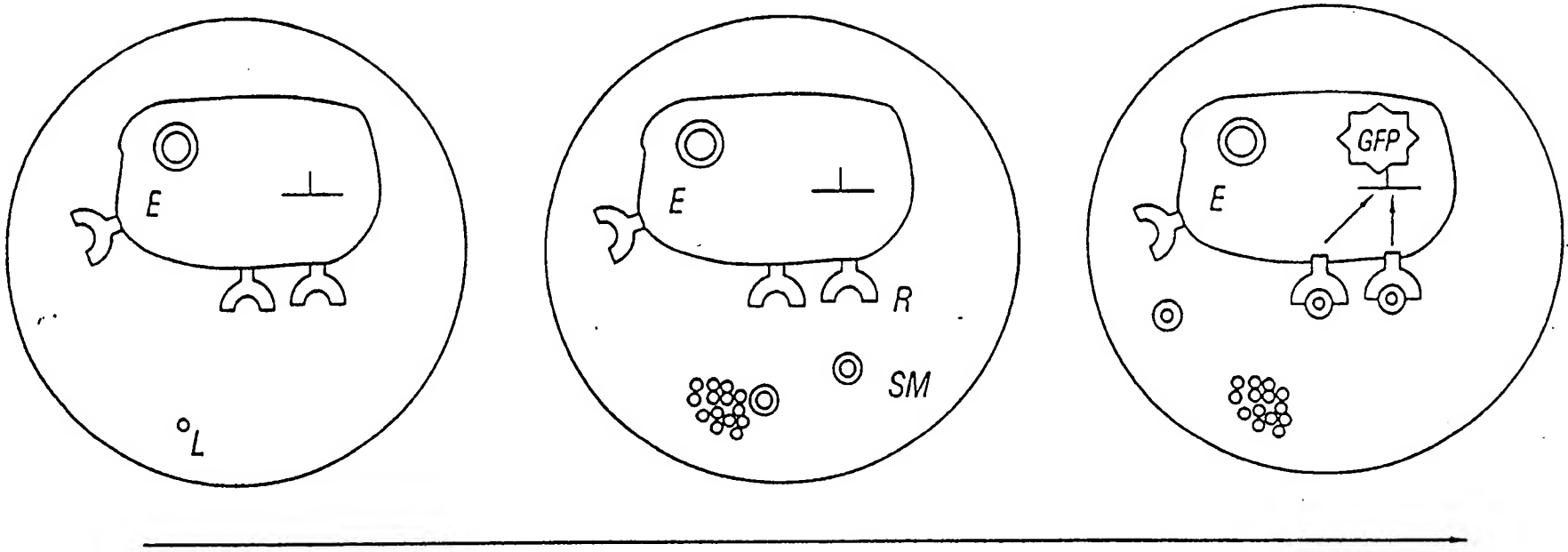


FIG. 2



Co-encapsulation Library + Eukaryote

Growth and expression of small molecule from host

Receptor binding of small molecule & GFP reporting

E=Eukaryotic assay organism	*L=Large insert library*	*SM=Small molecule*
GFP= Green fluorescent protein	*R=Eukaryotic receptor*	

FIG. 3

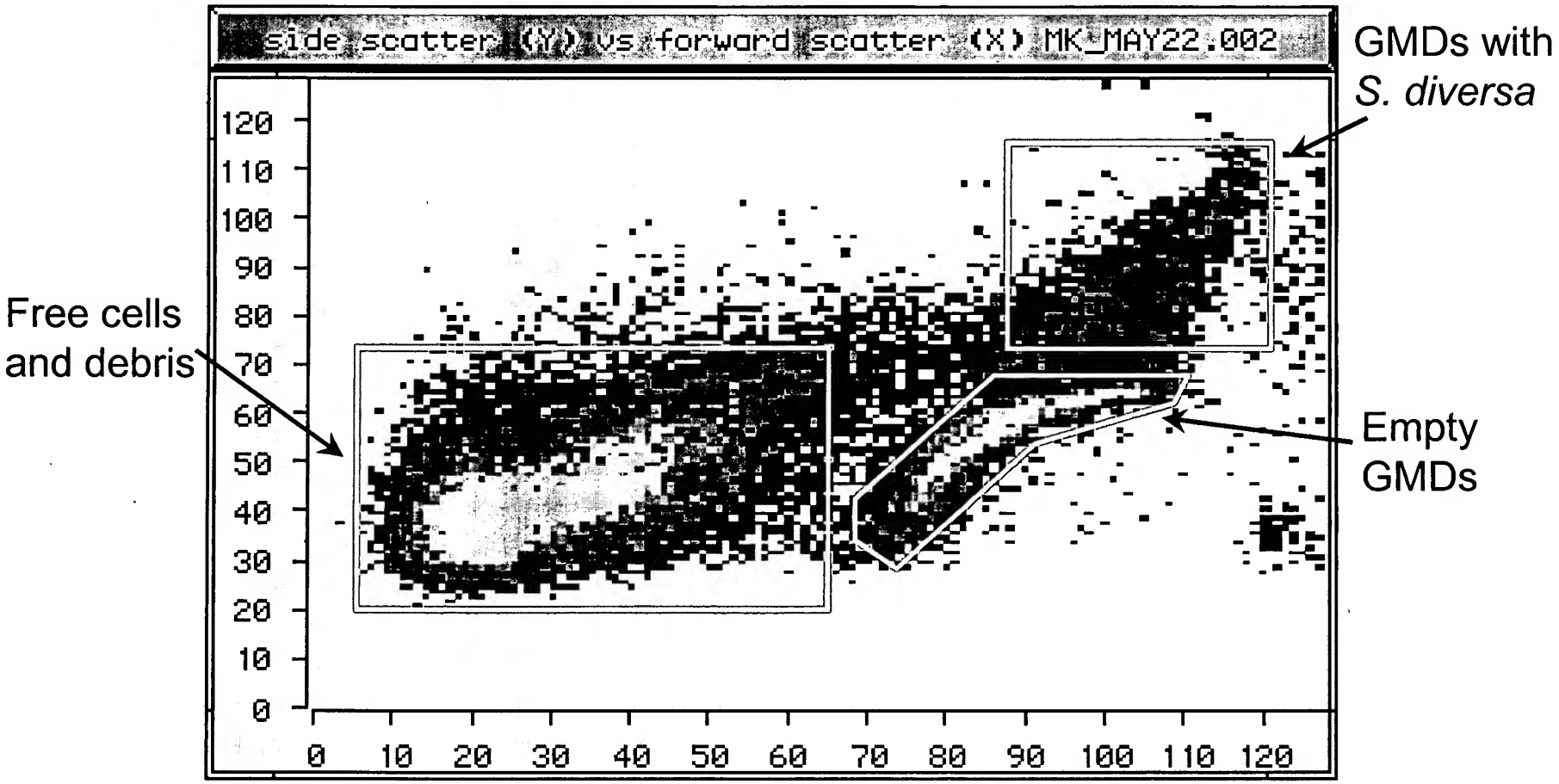

side scatter (Y) vs forward scatter (X) MK_MAY22.002
GMDs with
S. diversa
Free cells
and debris
Empty
GMDs
120
110
100
90
80
70
60
50
40
30
20
10
0
0
10
20
30
40
50
60
70
80
90
100
110
120

Fig 4

Whole Cell Hybridization Protocol
(+ control)

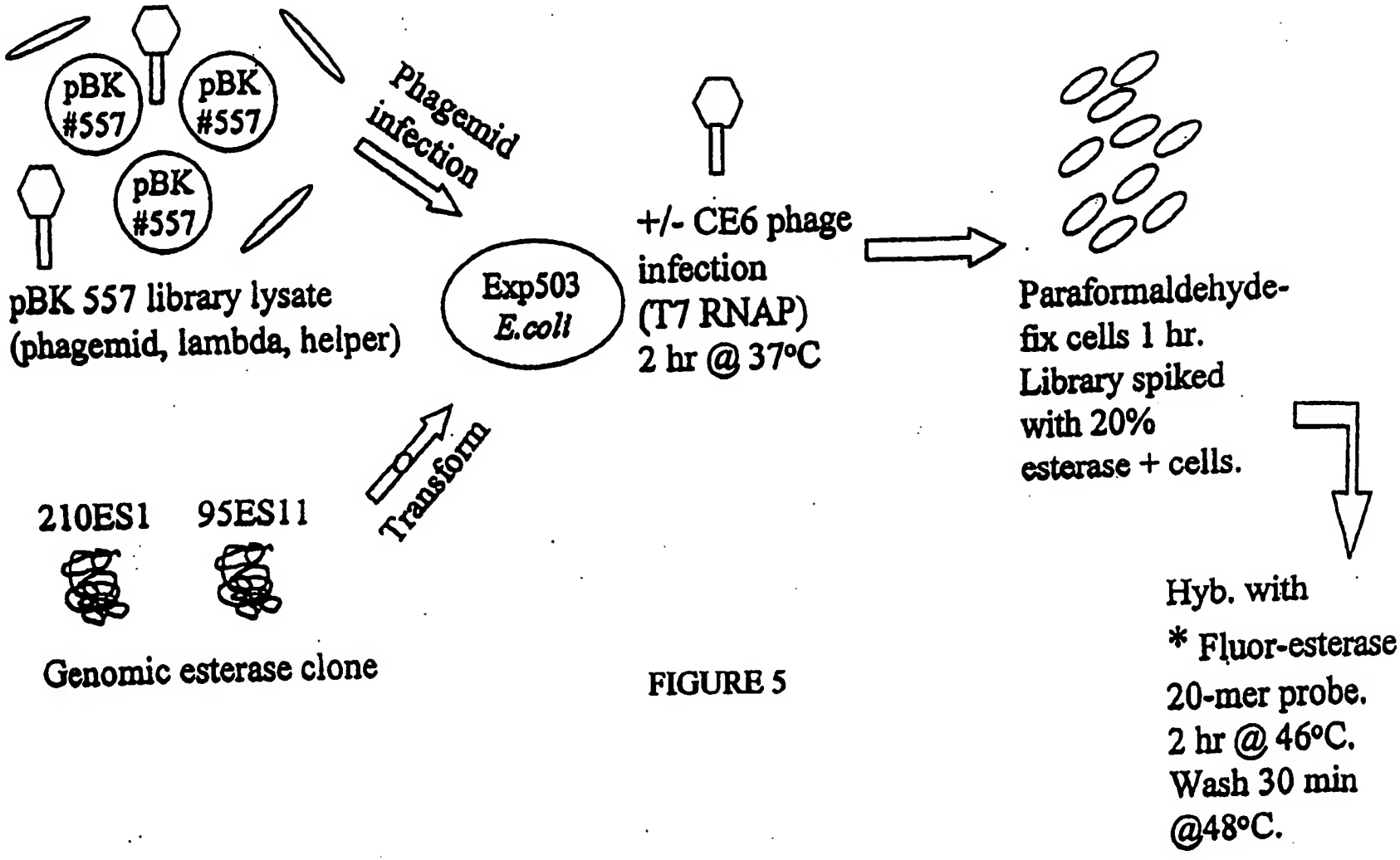


FIGURE 5

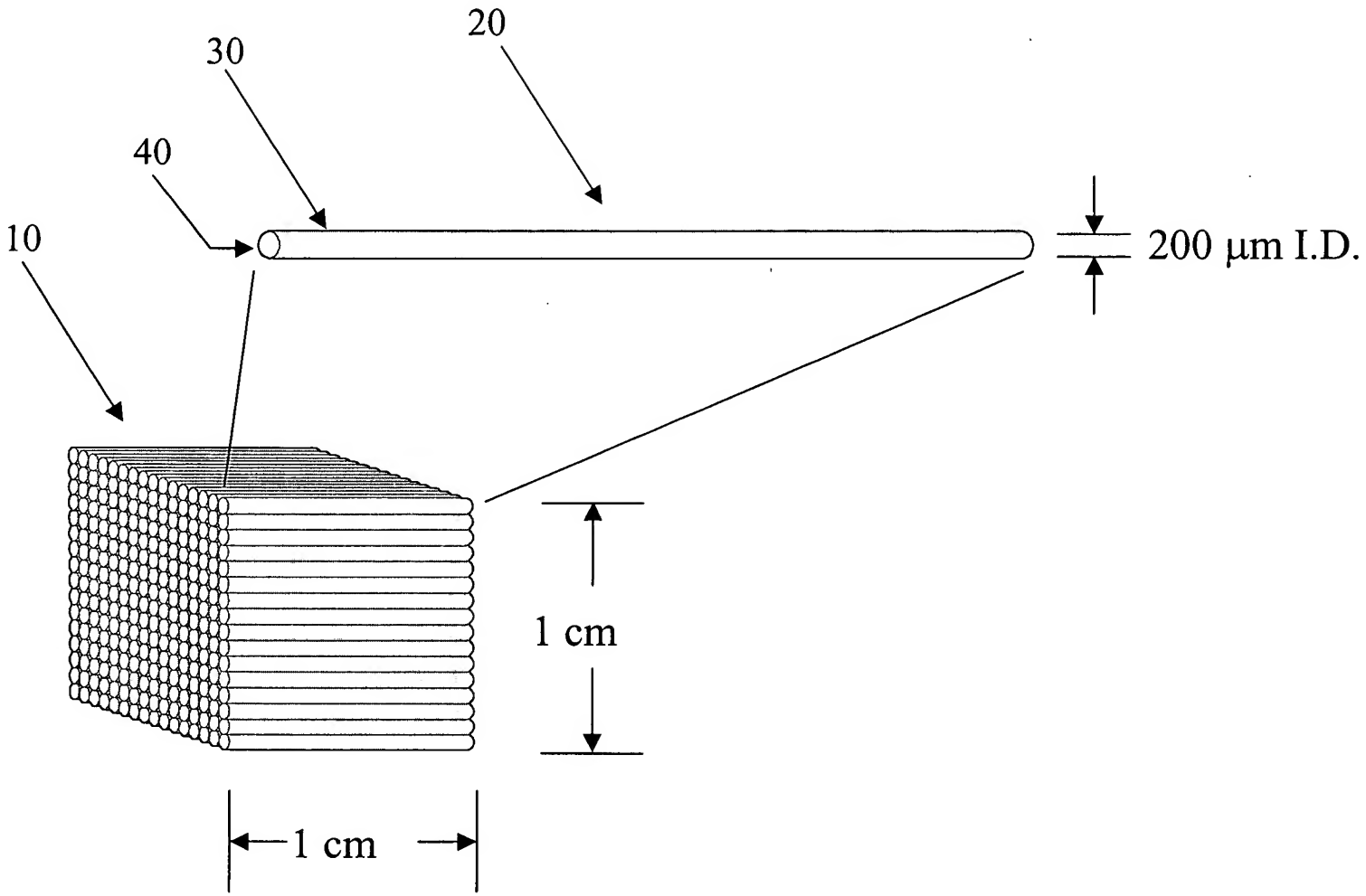


Fig. 6A

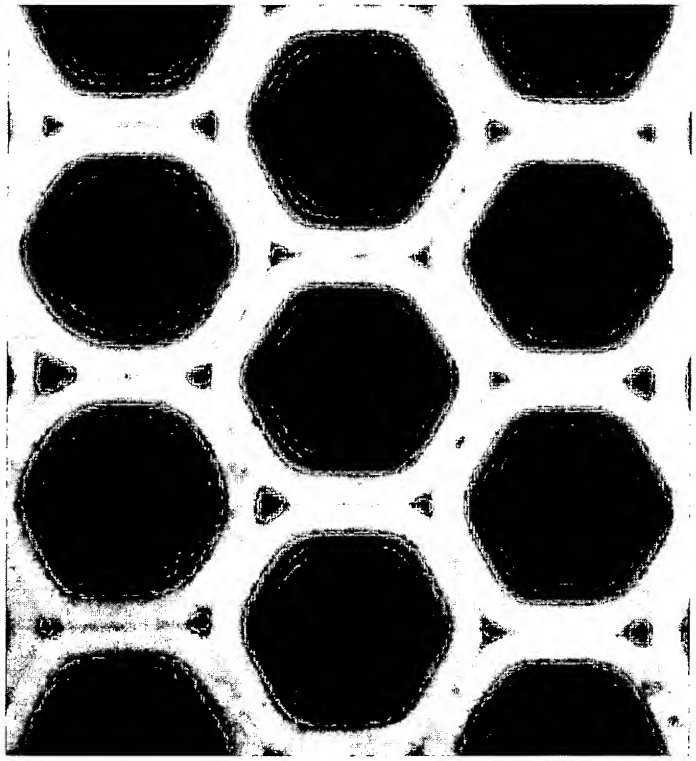


Fig. 6B

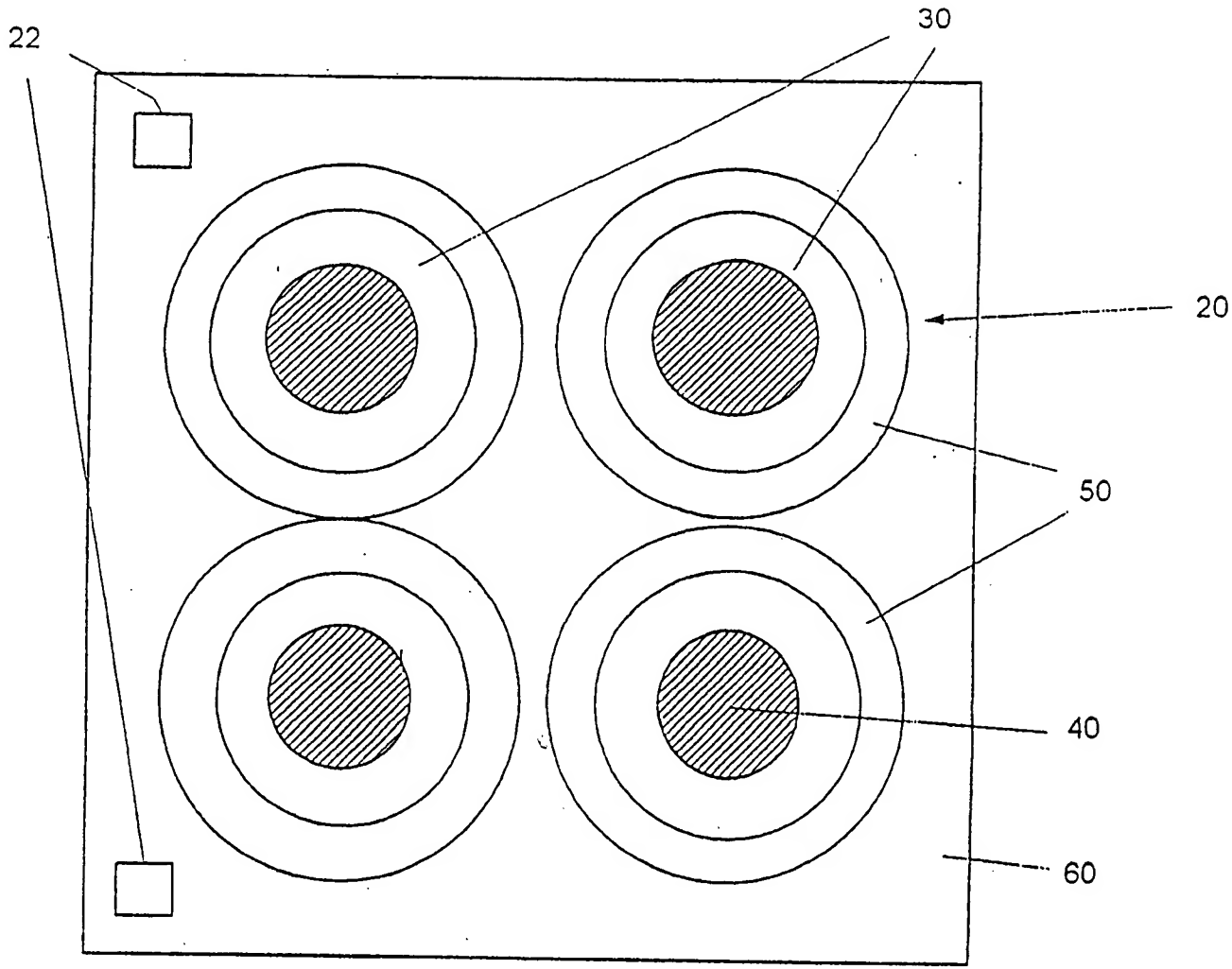


FIG. 7

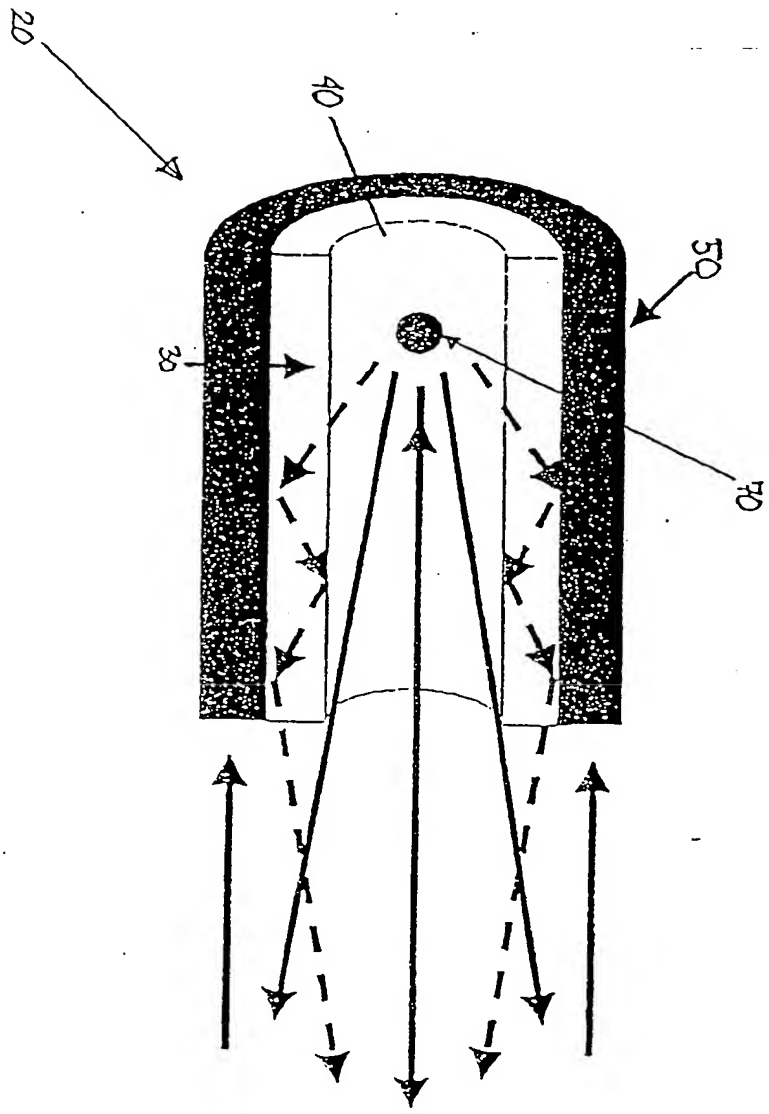


Figure 8

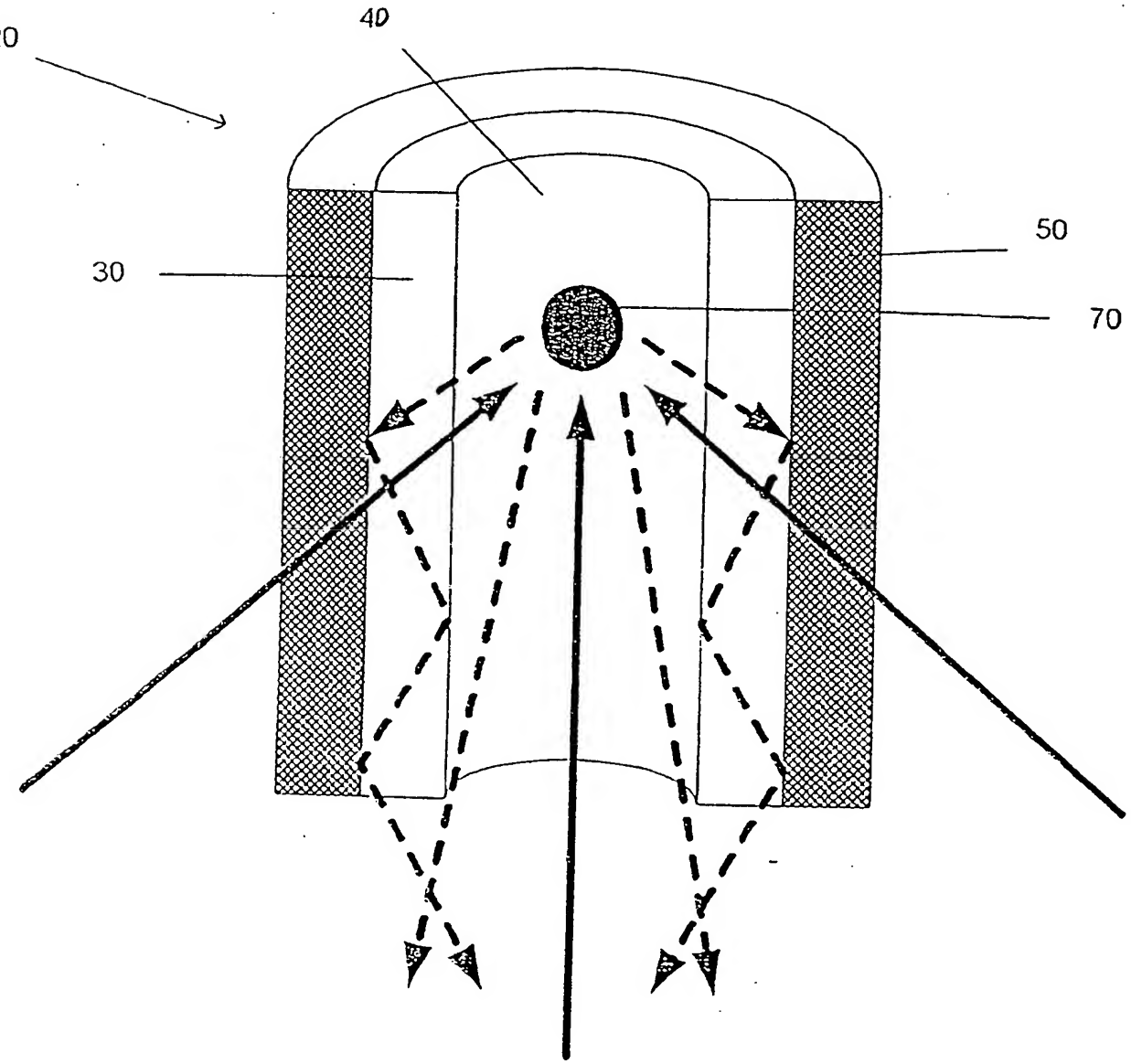


Figure 9

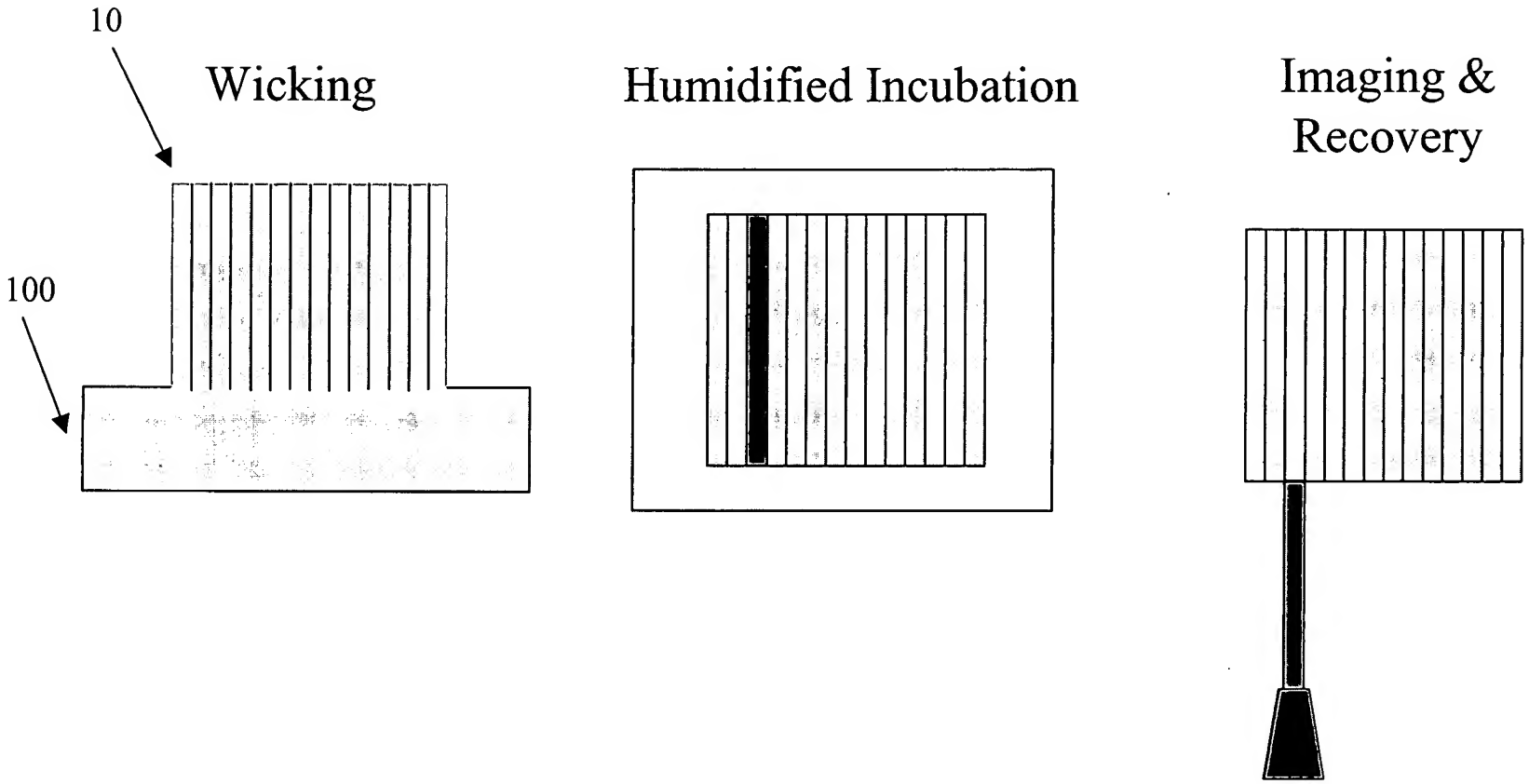


Fig. 10

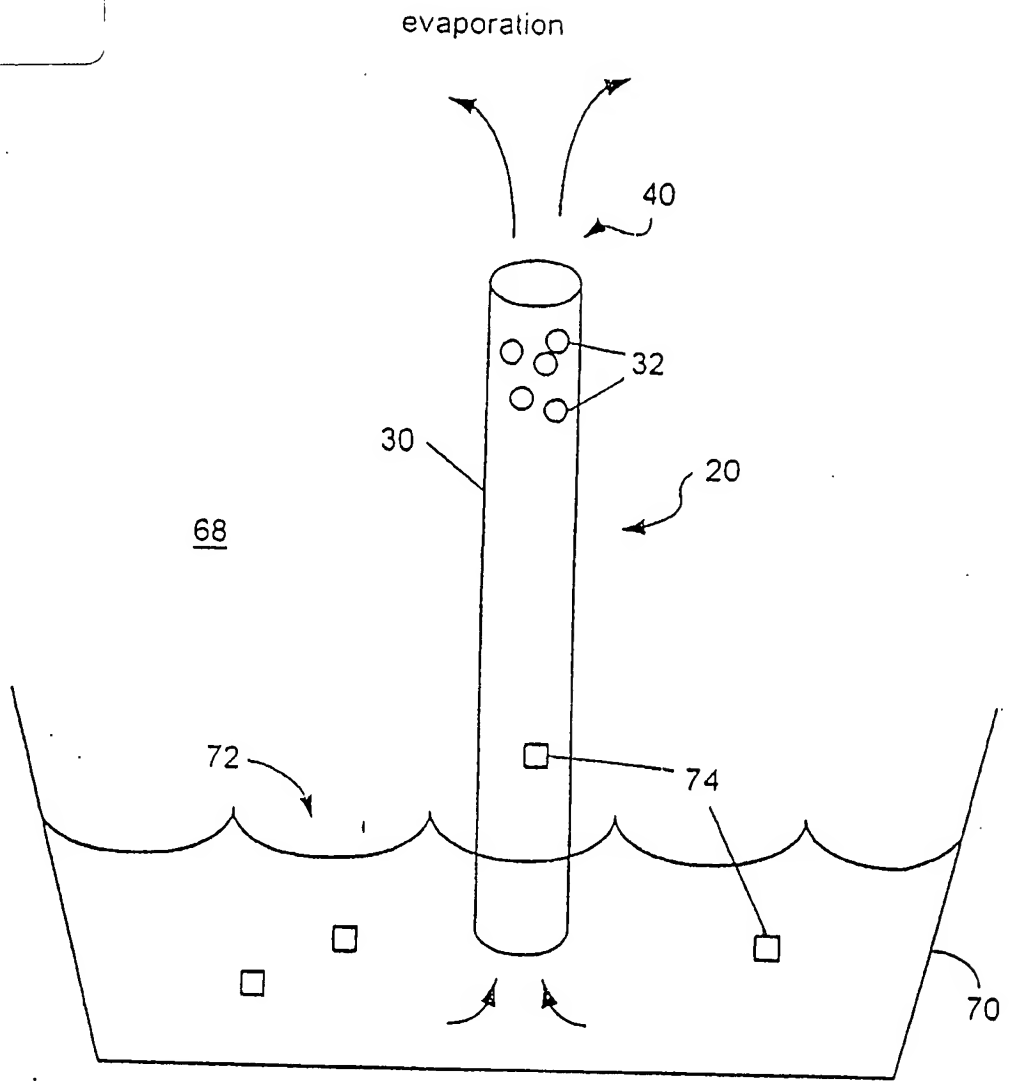


Figure 11

Figure 12A

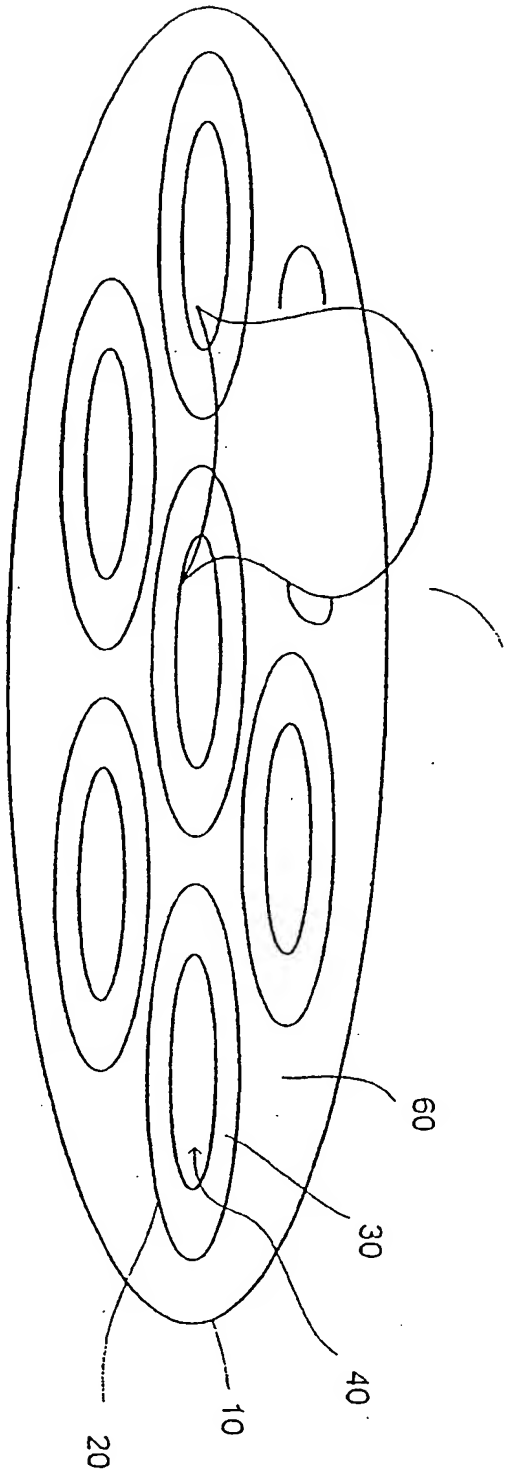
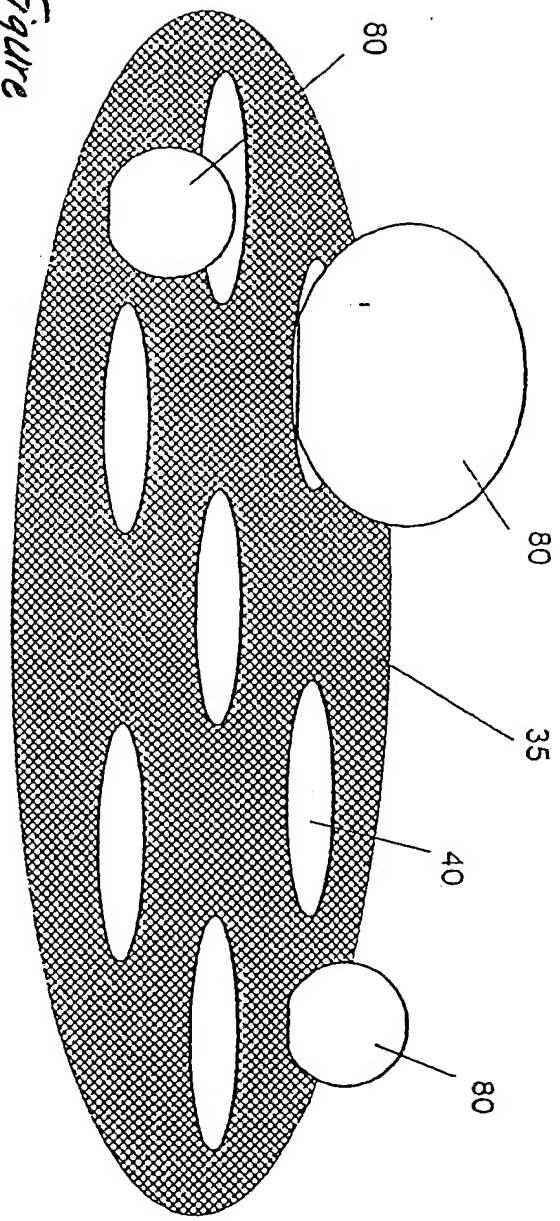


Figure 12B



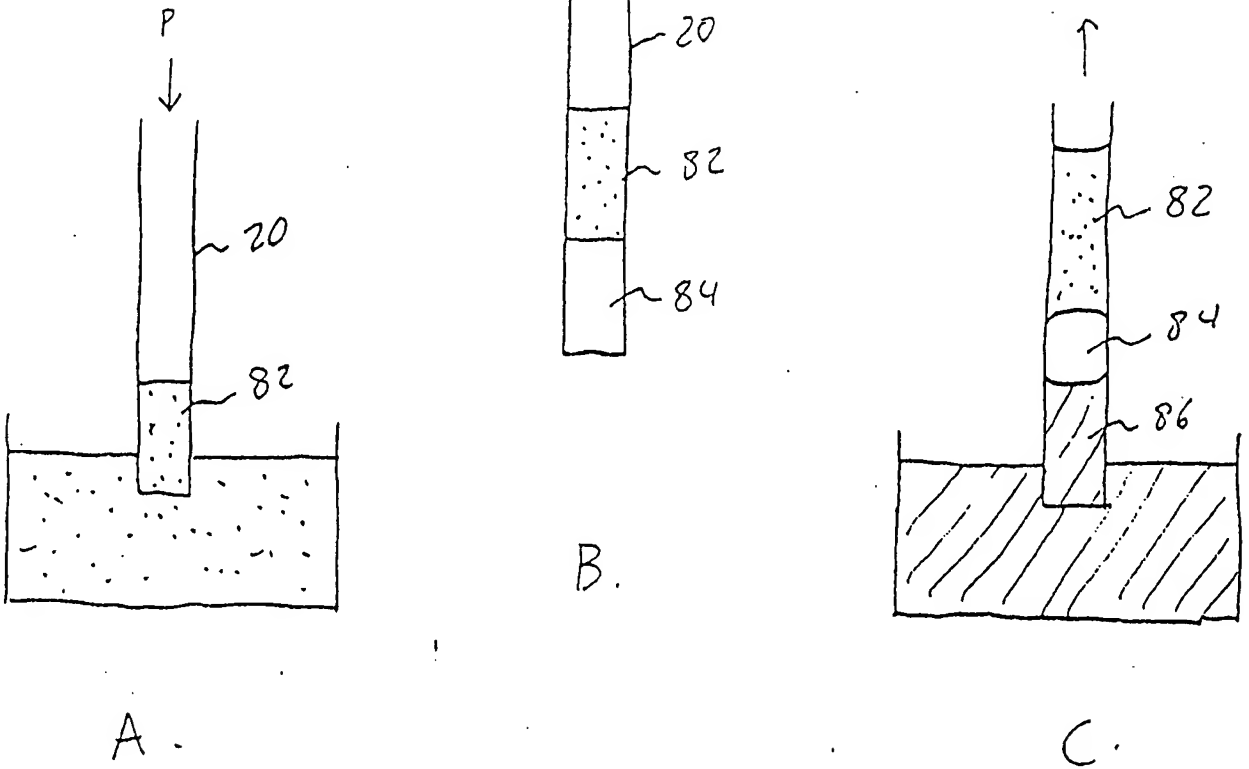


Figure 13

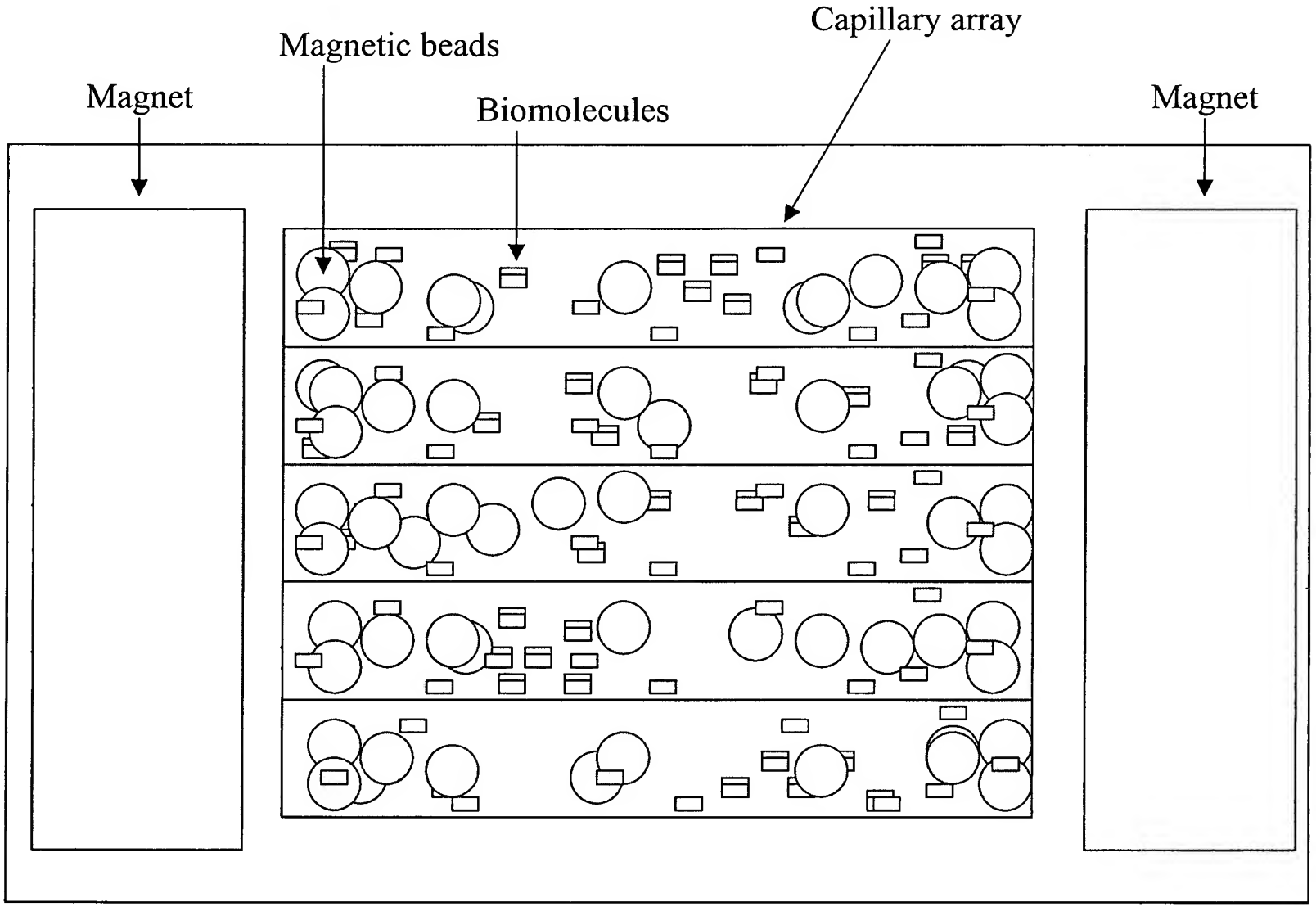

Capillary array
Magnetic beads
Magnet
Biomolecules
Magnet

Fig. 14A

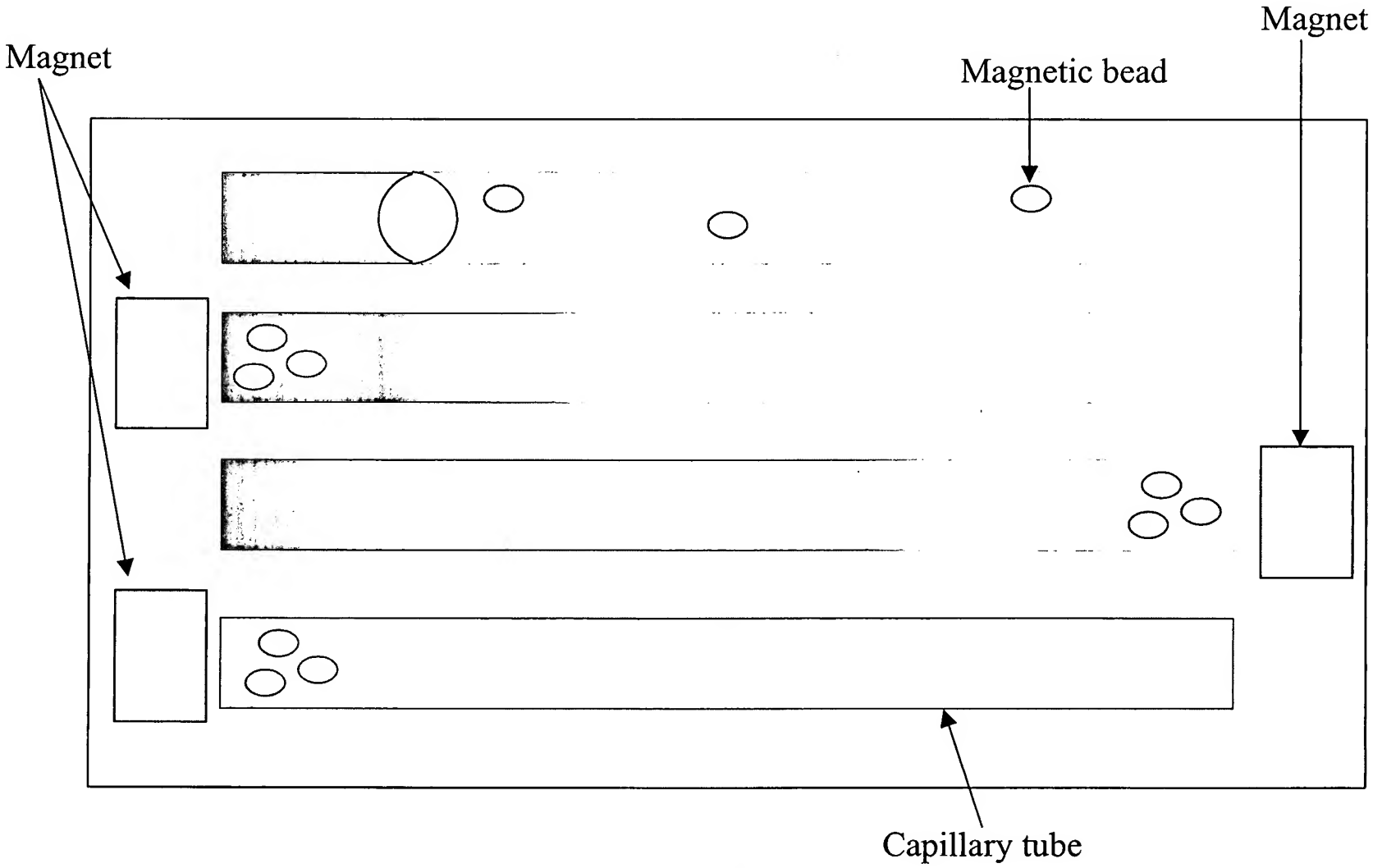


Fig. 14B

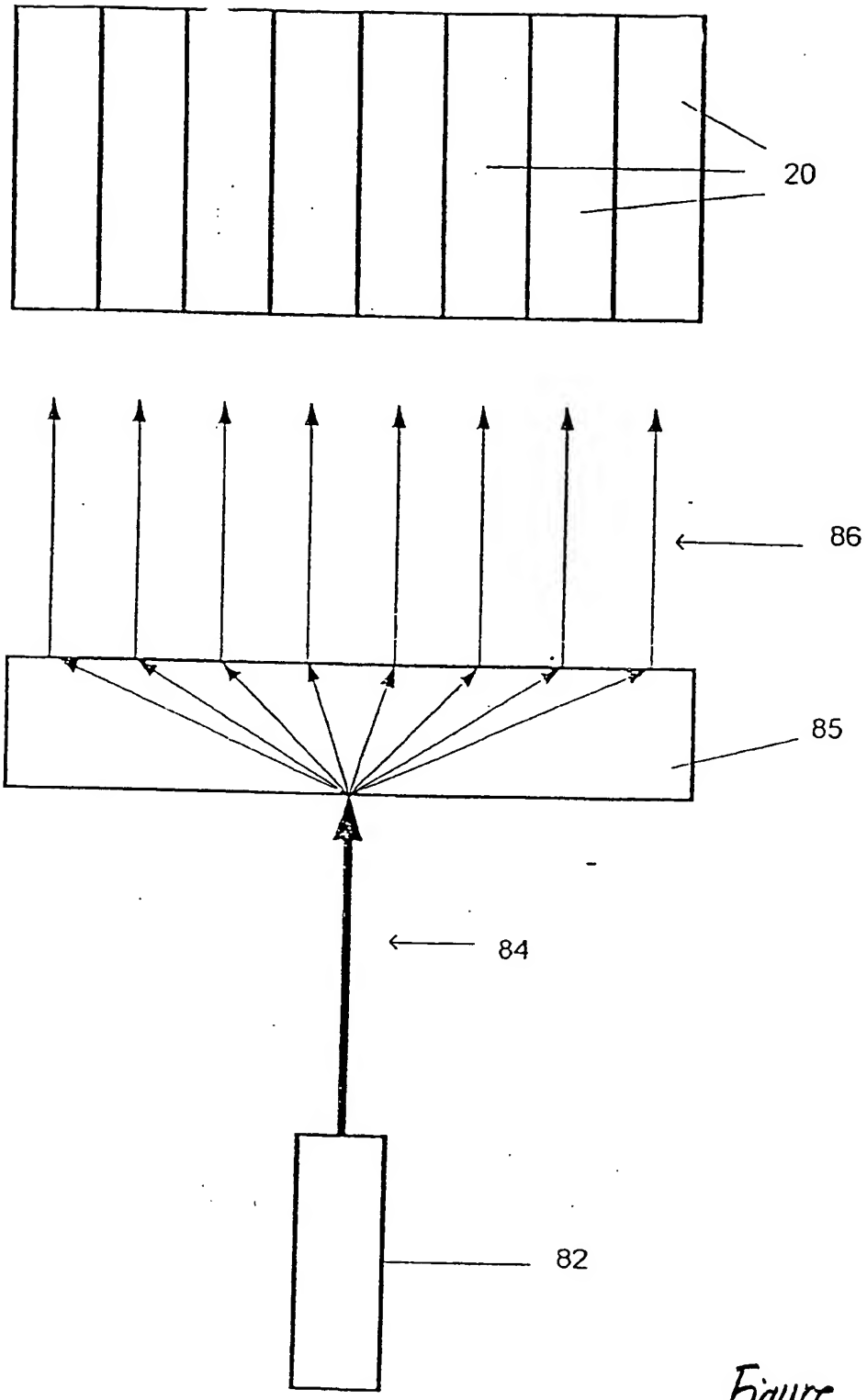
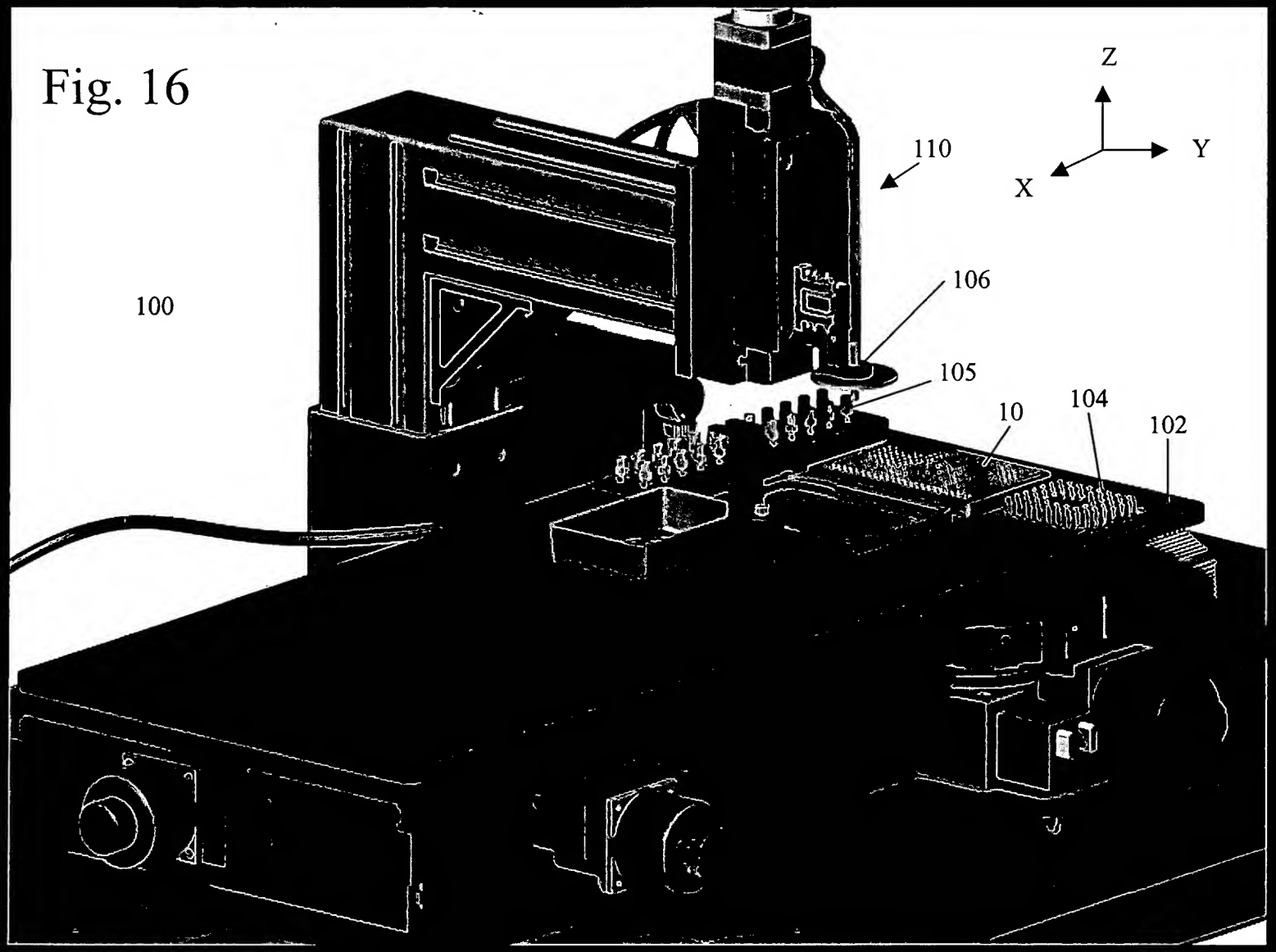


Figure 15

Automated Hit Detection & Recovery System

Fig. 16



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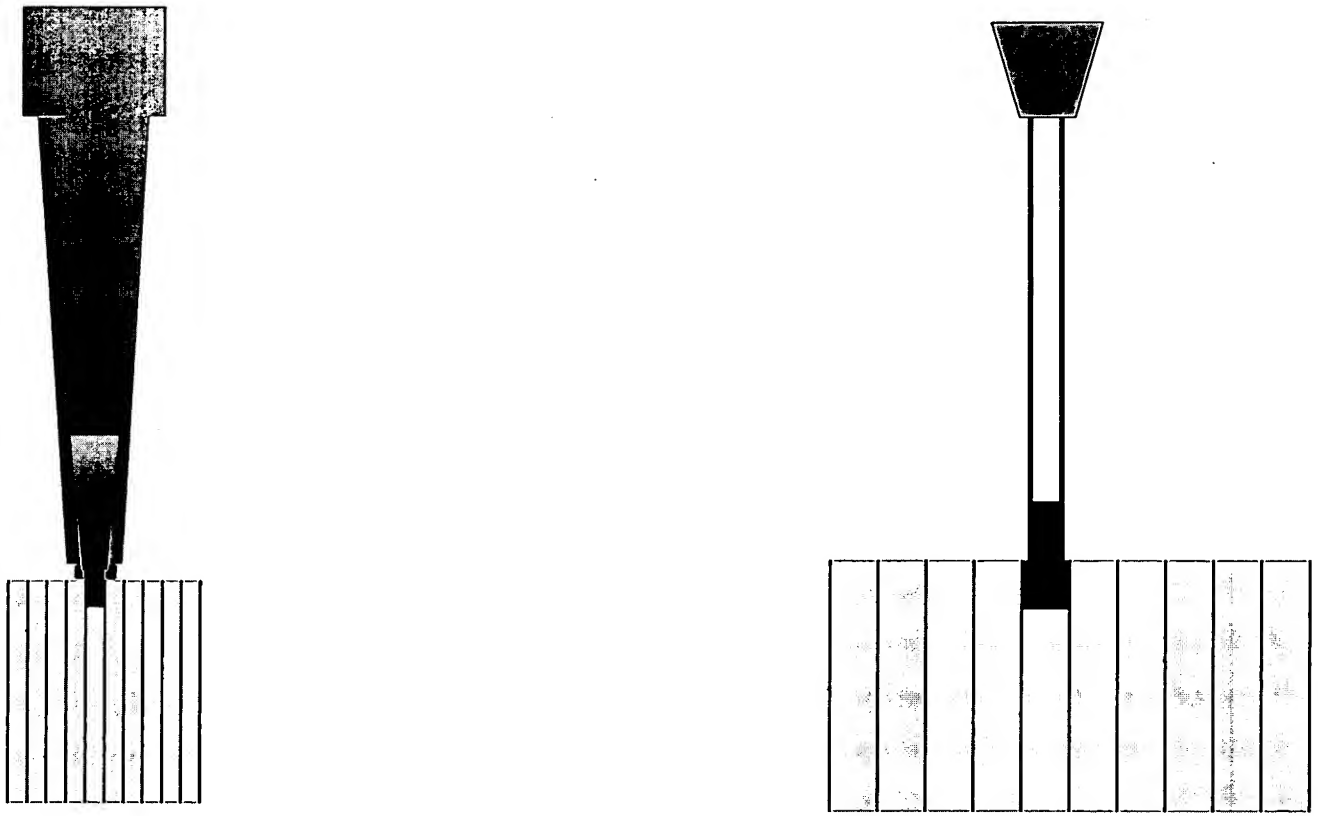


Fig. 17

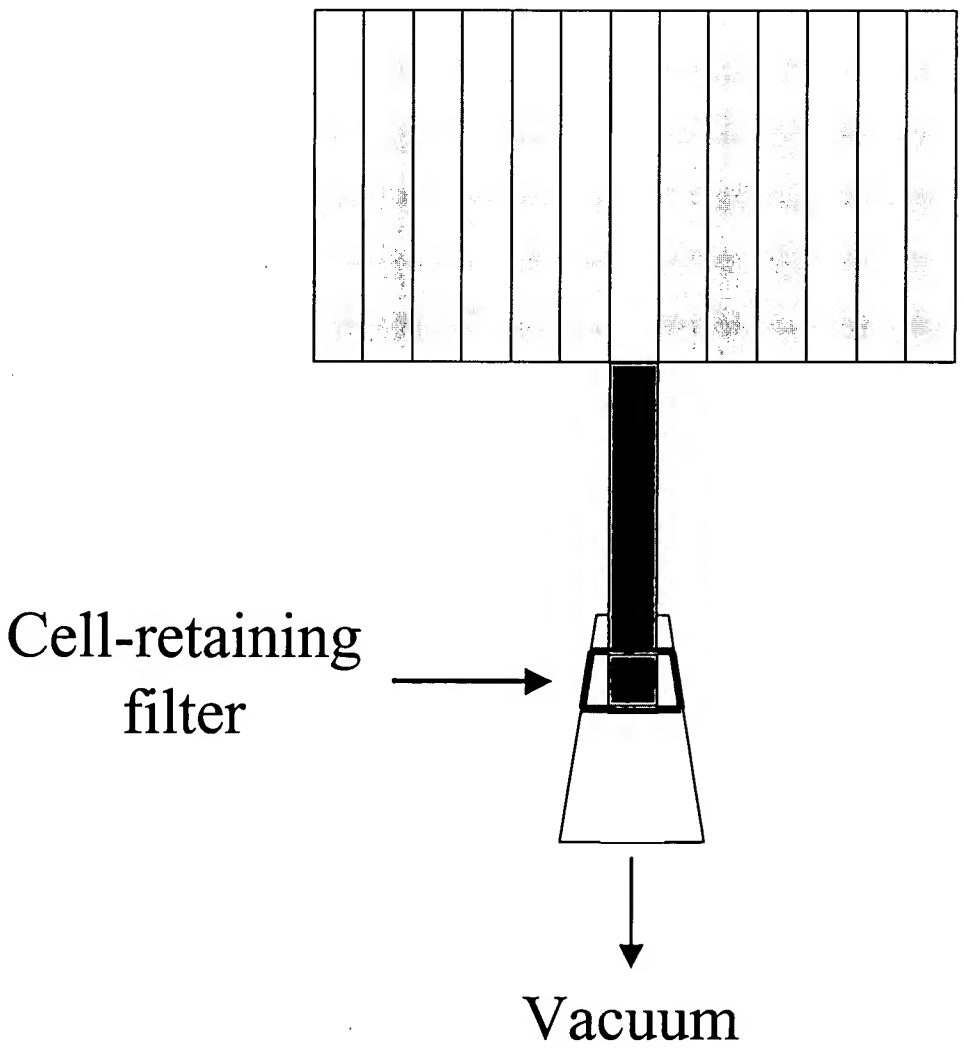


Fig. 17C

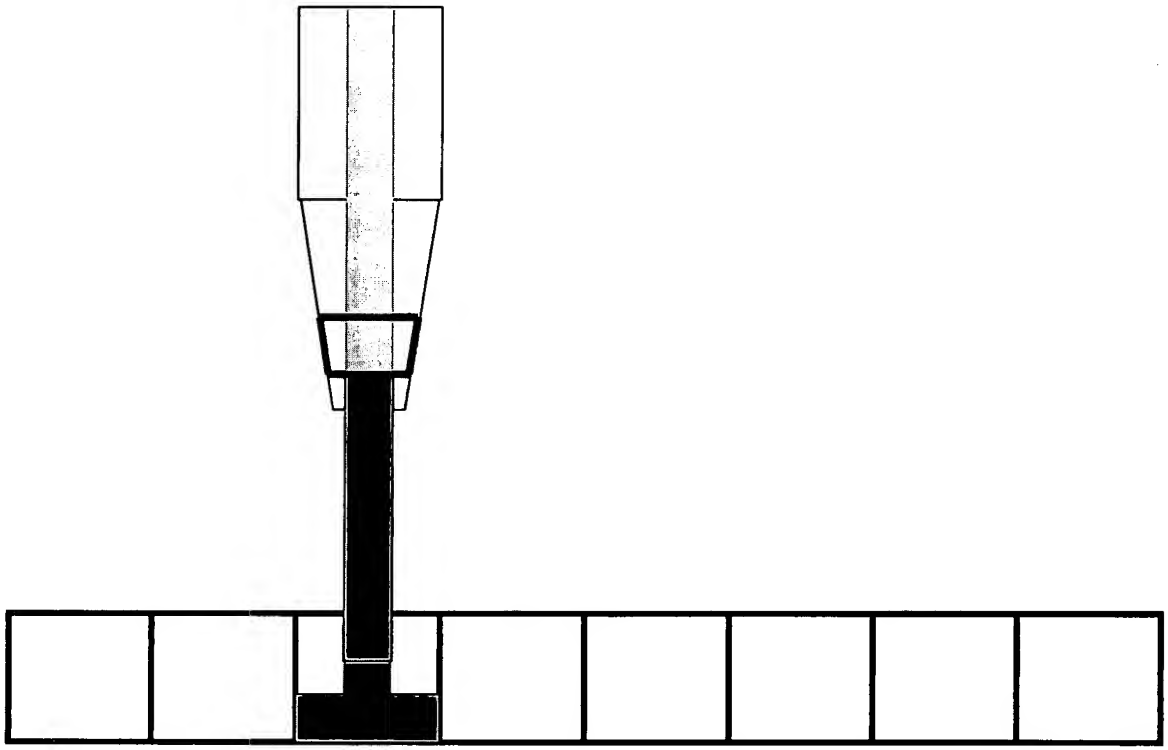


Fig. 17D

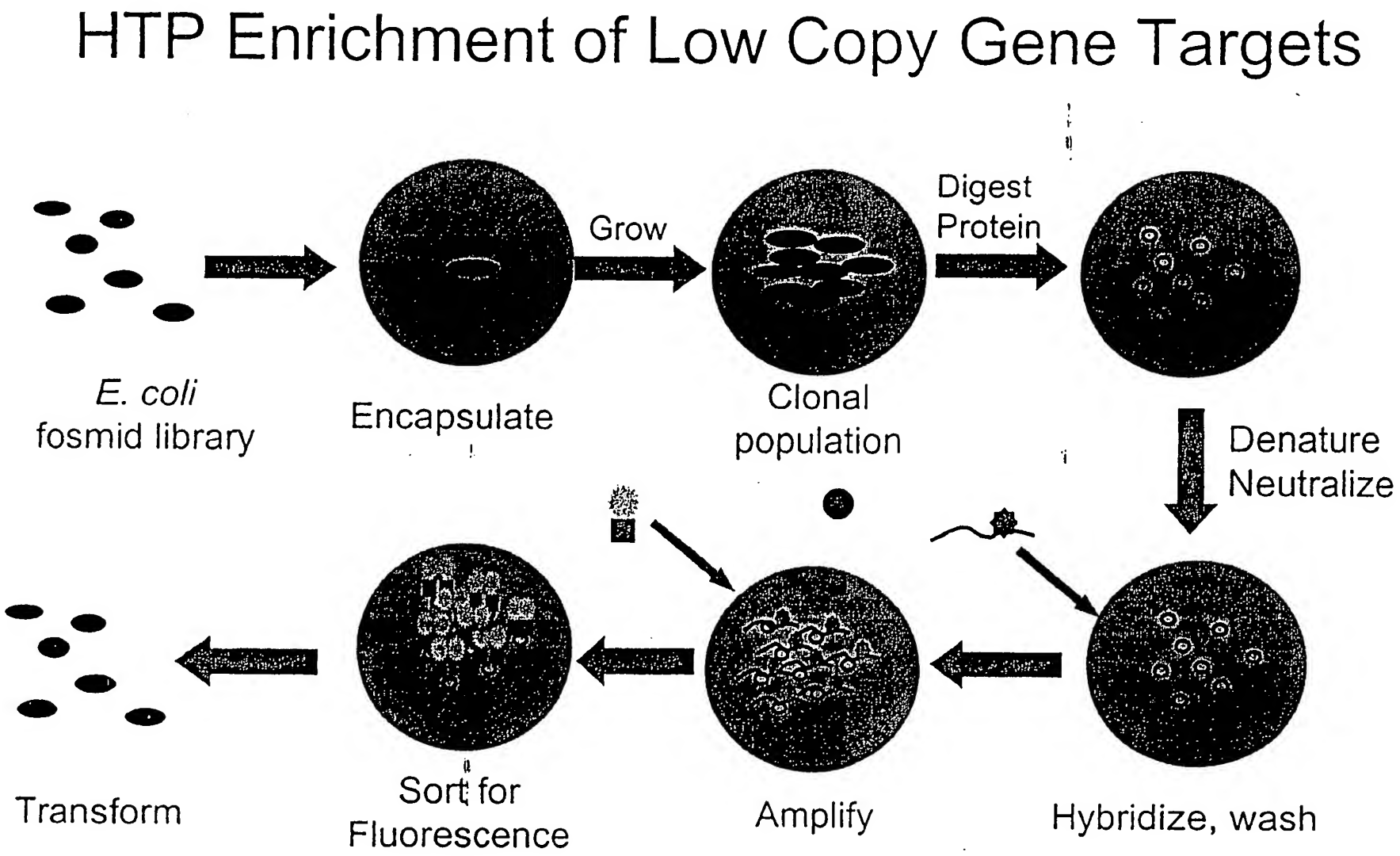
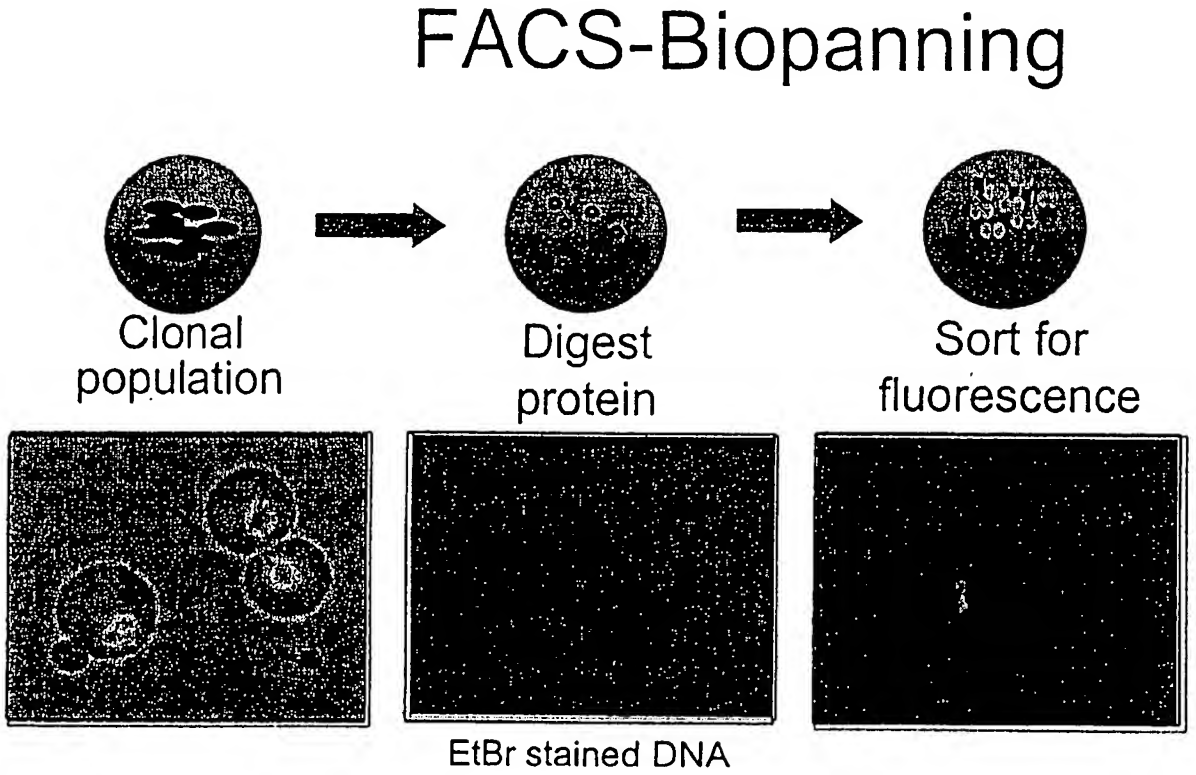

HTP Enrichment of Low Copy Gene Targets
E. coli
fosmid library
Encapsulate
Grow
Clonal
population
Digest
Protein
Denature
Neutralize
Hybridize, wash
Amplify
Sort for
Fluorescence
Transform

FIG. 18

FIG. 19



Alignment of Environmental PKS Sequence

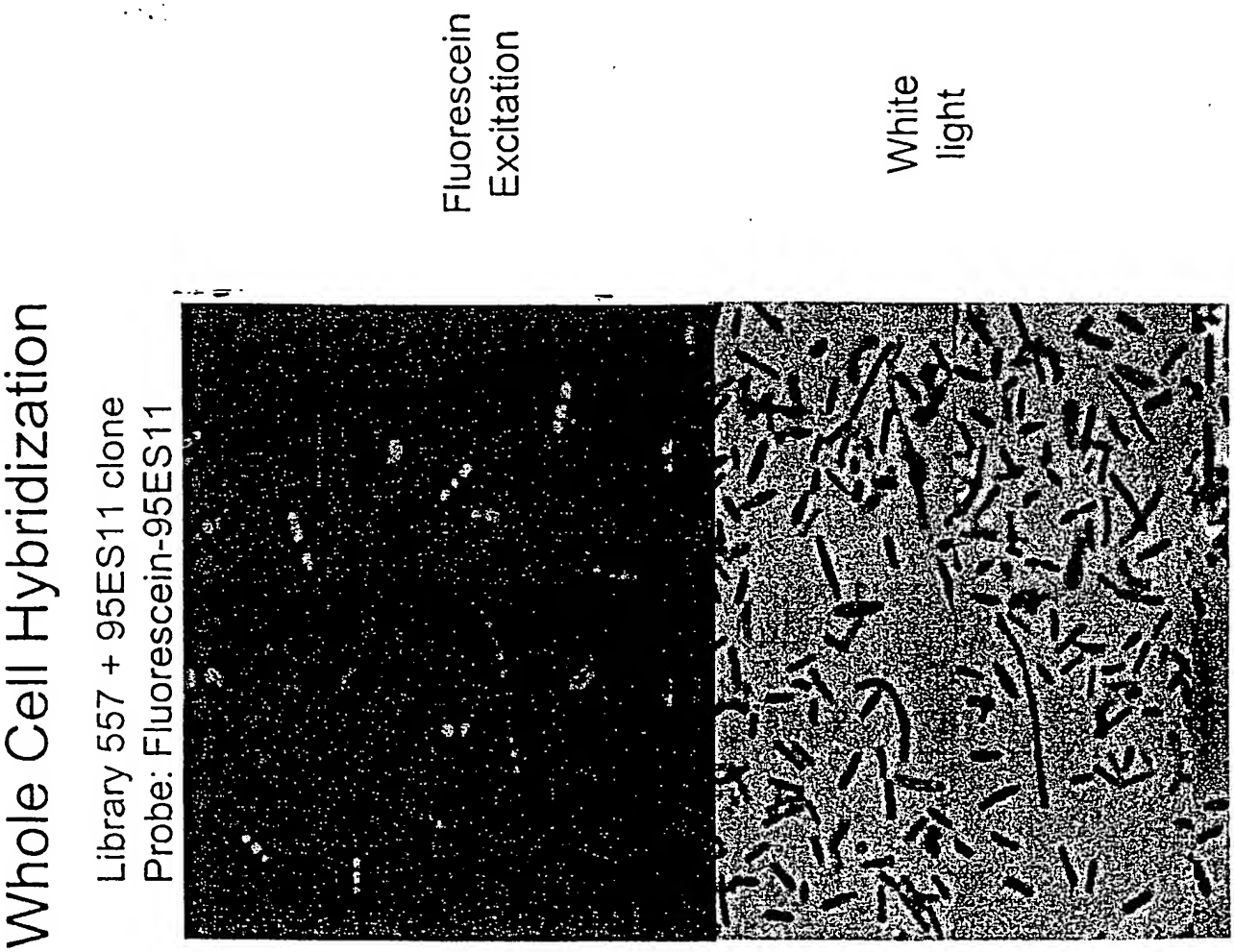

Whole Cell Hybridization
Library 557 + 95ES11 clone
Probe: Fluorescein-95ES11
Fluorescein Excitation
White light

FIG. 20

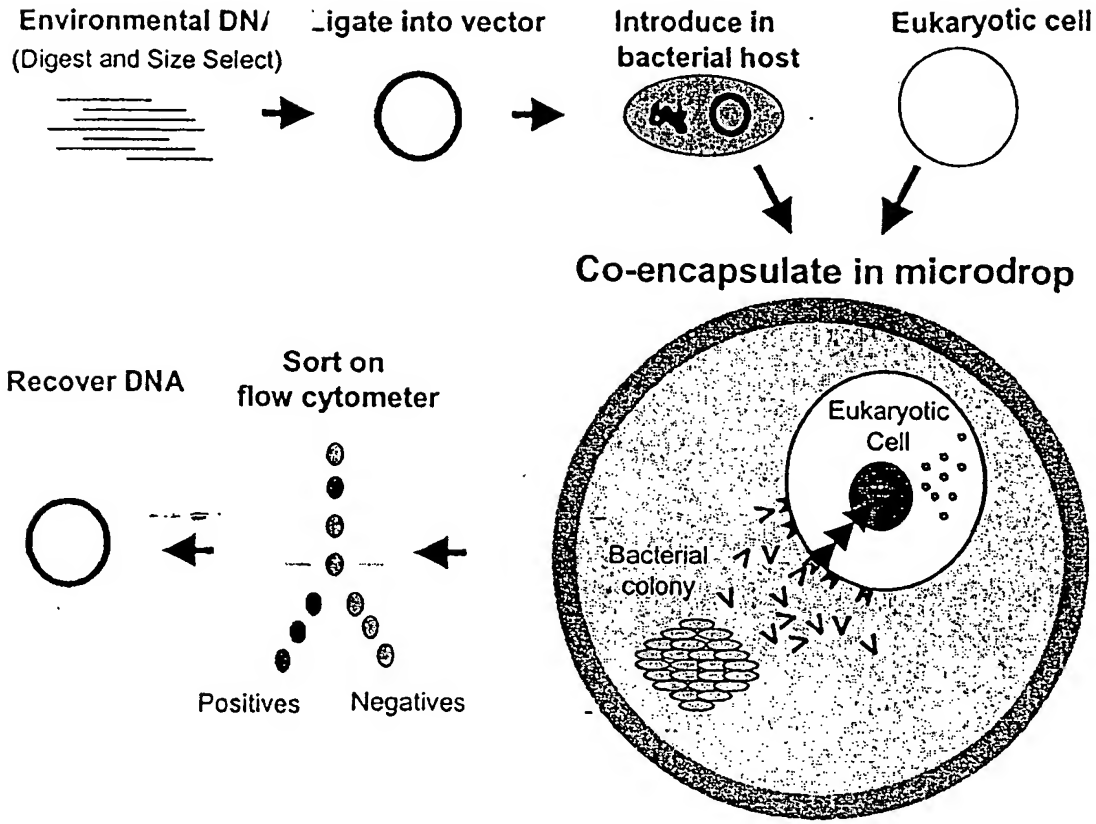

Environmental DNA
(Digest and Size Select)
Ligate into vector
Introduce in bacterial host
Eukaryotic cell
Co-encapsulate in microdrop
Eukaryotic Cell
Bacterial colony
Recover DNA
Sort on flow cytometer
Positives
Negatives

FIG. 21

Whole Cell Hybridization Protocol

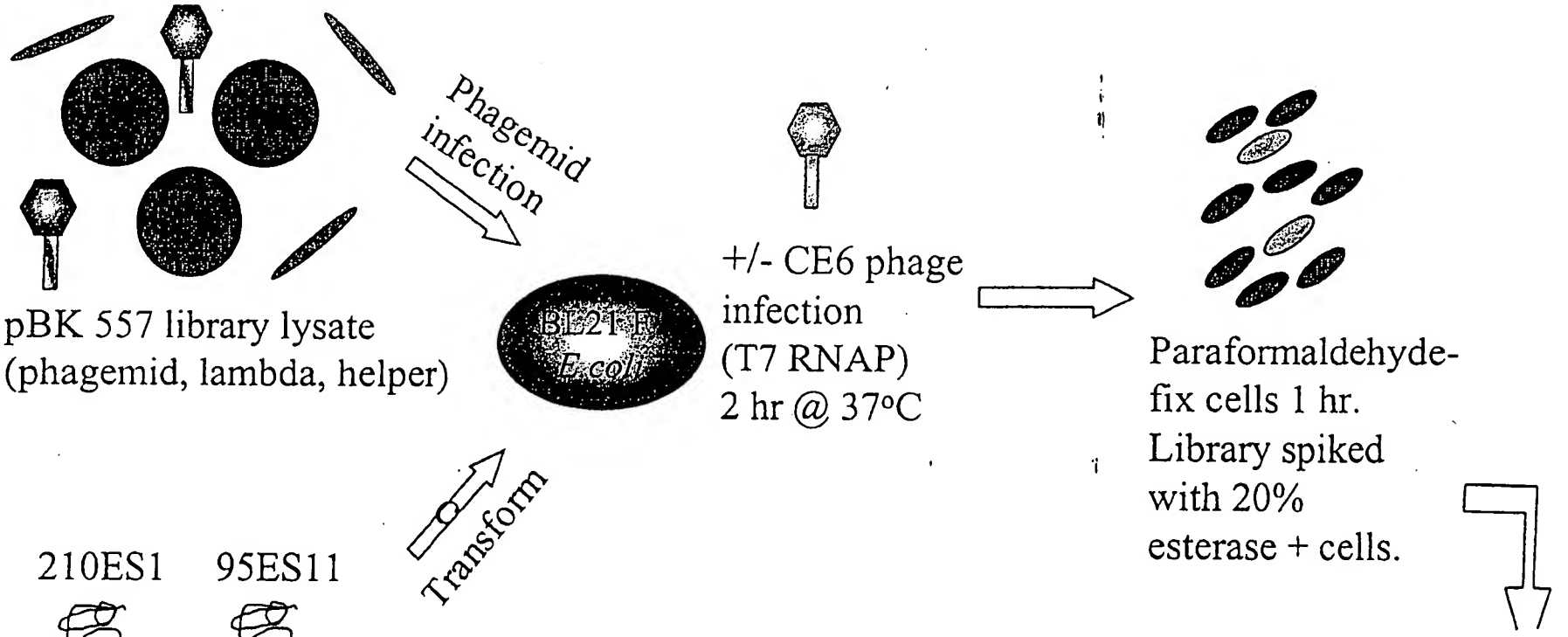


FIG. 22

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T7 RNA Polymerase Expression System

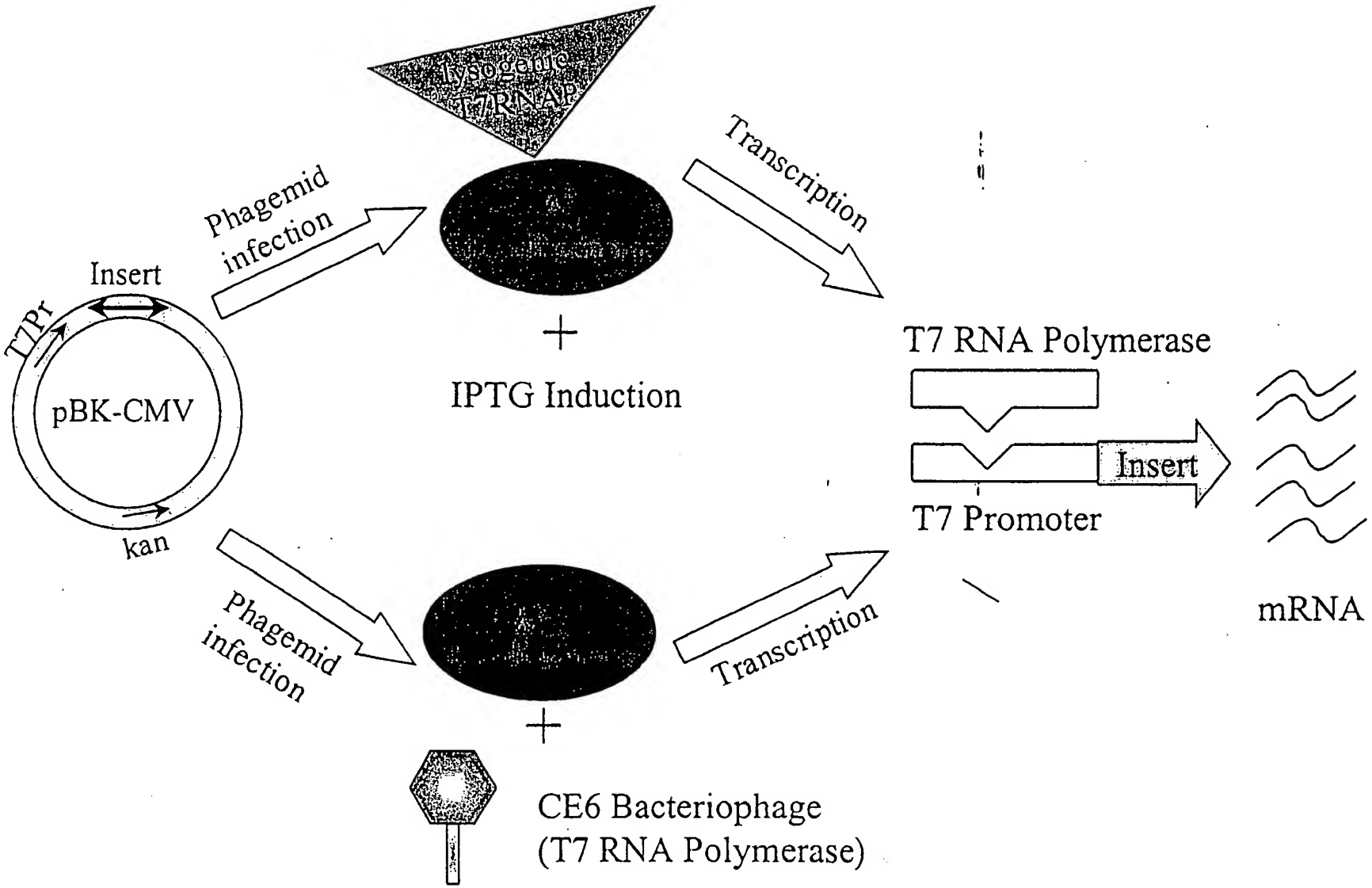

FIG. 23

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Title: HIGH THROUGHPUT SCREENING OF LIBRARIES

Figure 24

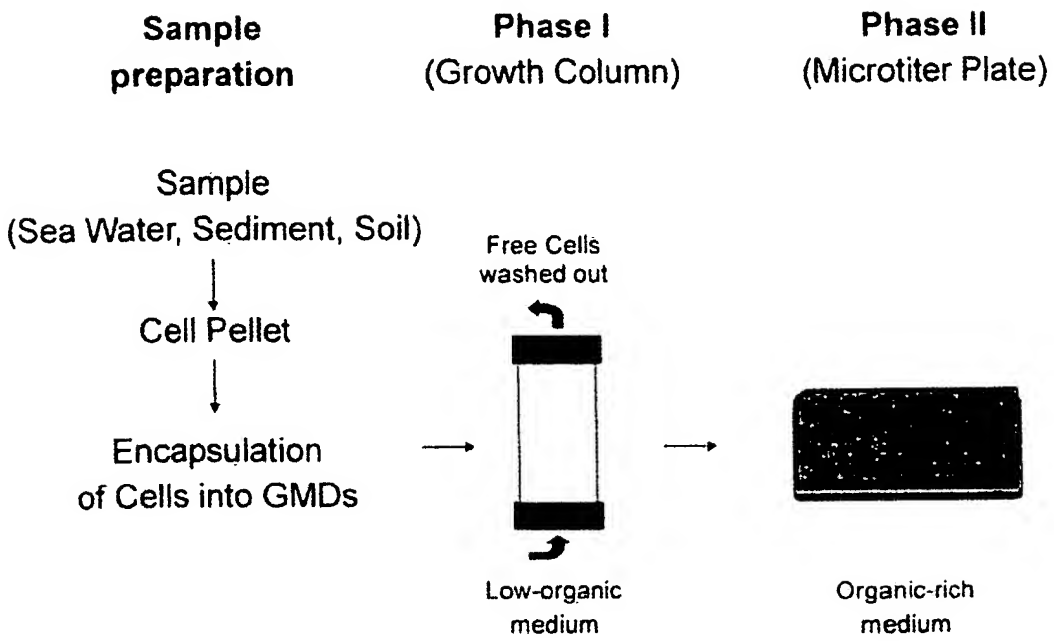


Fig. 24

Figure 25

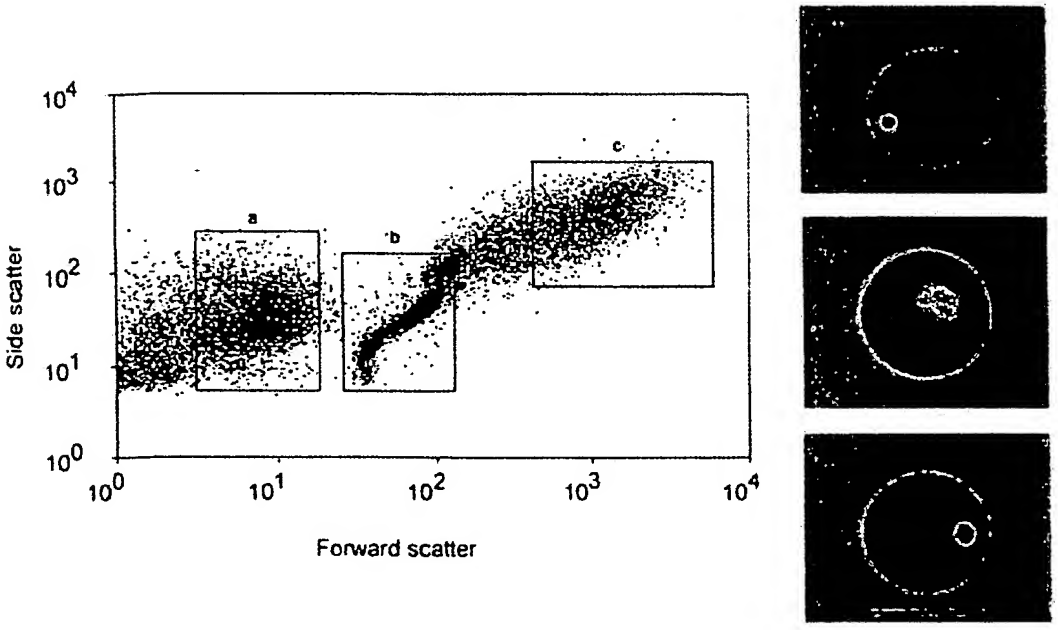


Fig. 25

Figure 26

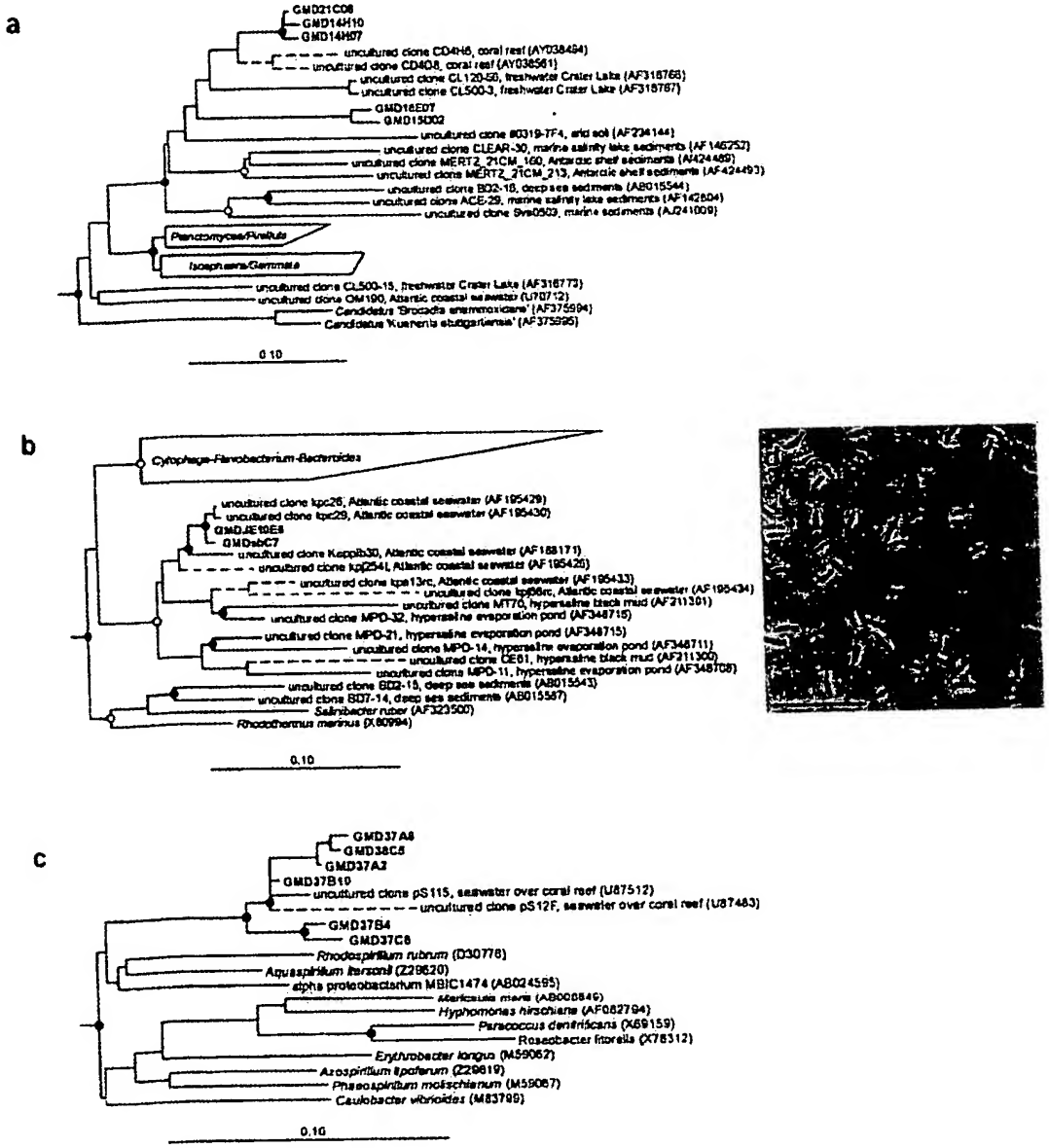
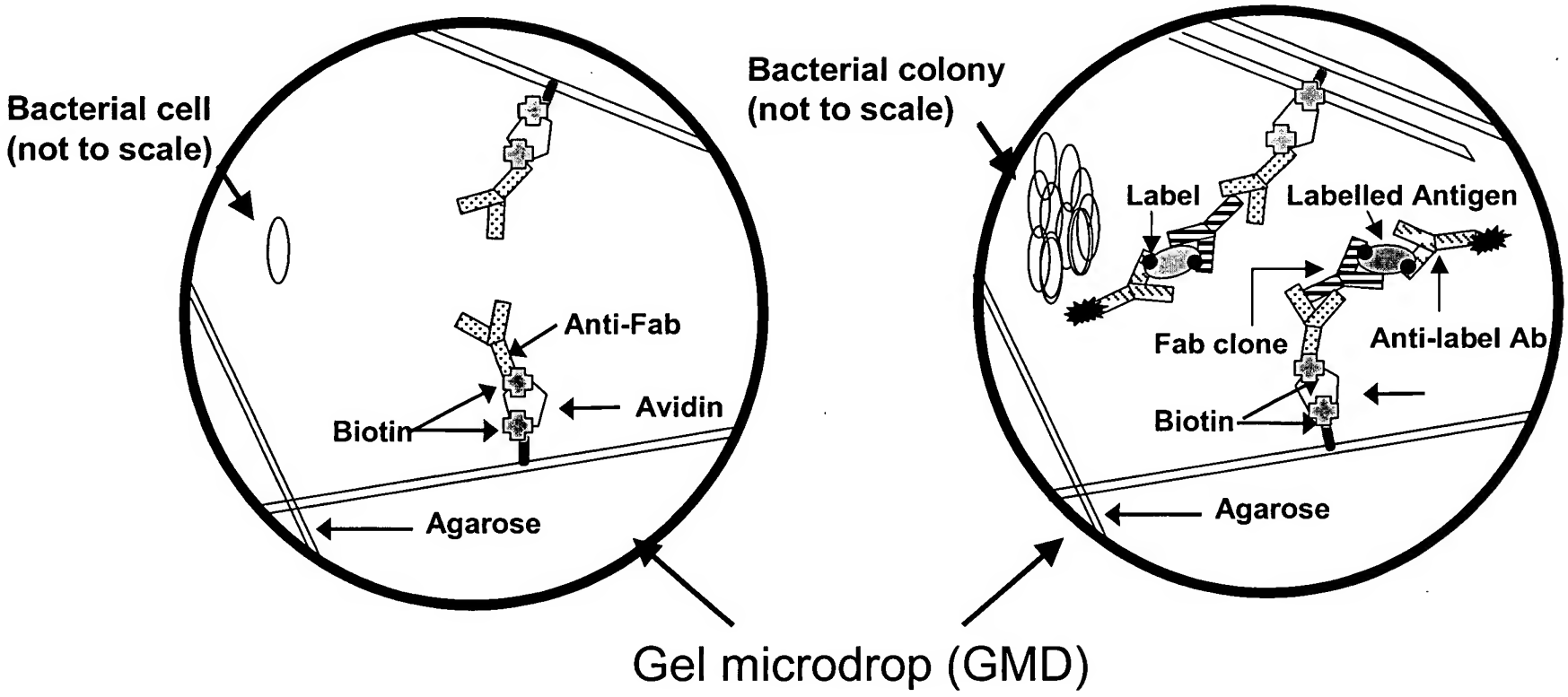


Fig. 26

Fig. 27


Bacterial cell
(not to scale)
Anti-Fab
Avidin
Biotin
Agarose
Bacterial colony
(not to scale)
Label
Labelled Antigen
Fab clone
Anti-label Ab
Biotin
Agarose
Gel microdrop (GMD)

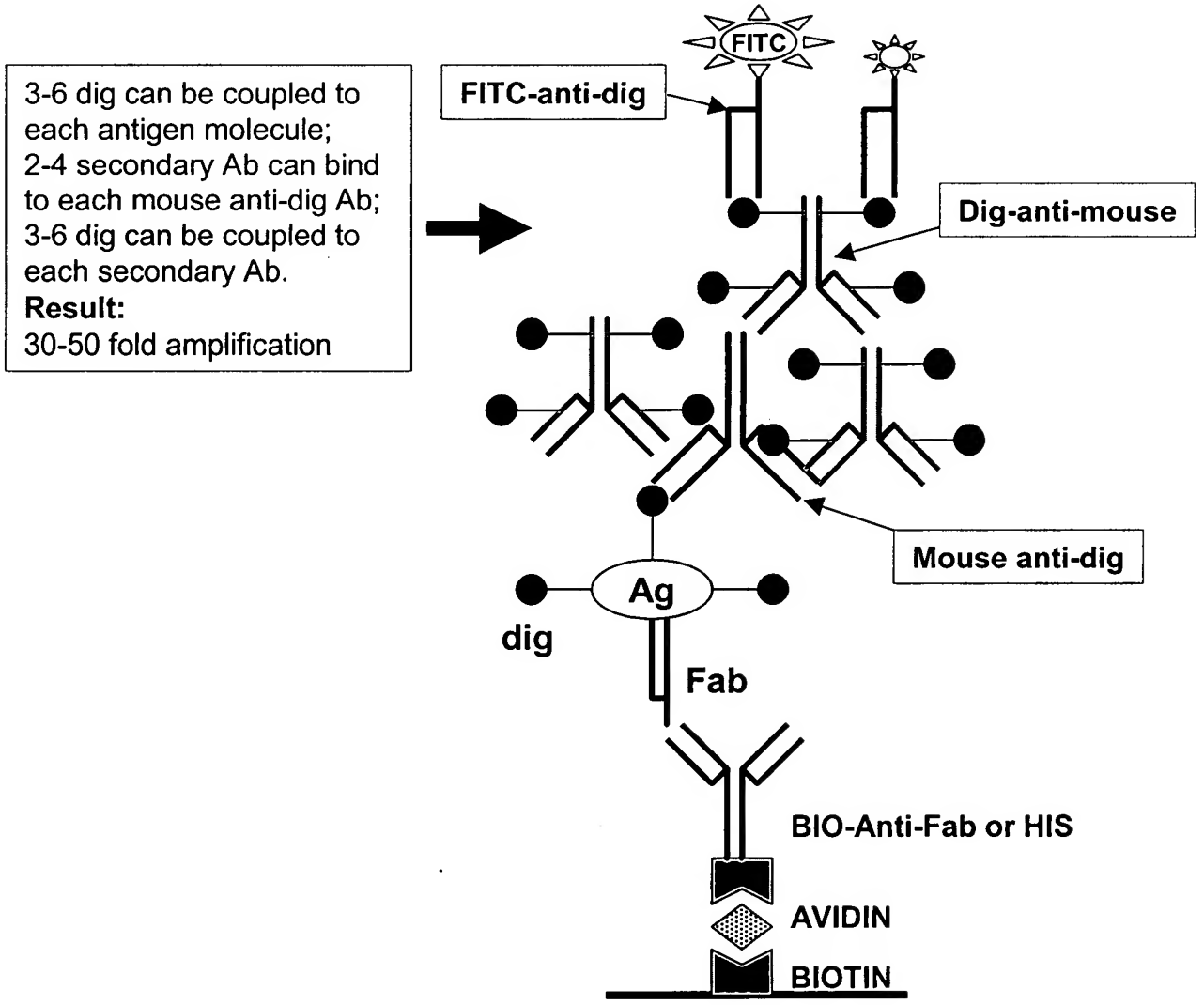

3-6 dig can be coupled to each antigen molecule;
2-4 secondary Ab can bind to each mouse anti-dig Ab;
3-6 dig can be coupled to each secondary Ab.
Result:
30-50 fold amplification
FITC
FITC-anti-dig
Dig-anti-mouse
Mouse anti-dig
Ag
dig
Fab
BIO-Anti-Fab or HIS
AVIDIN
BIOTIN

Fig. 28

Fig. 29

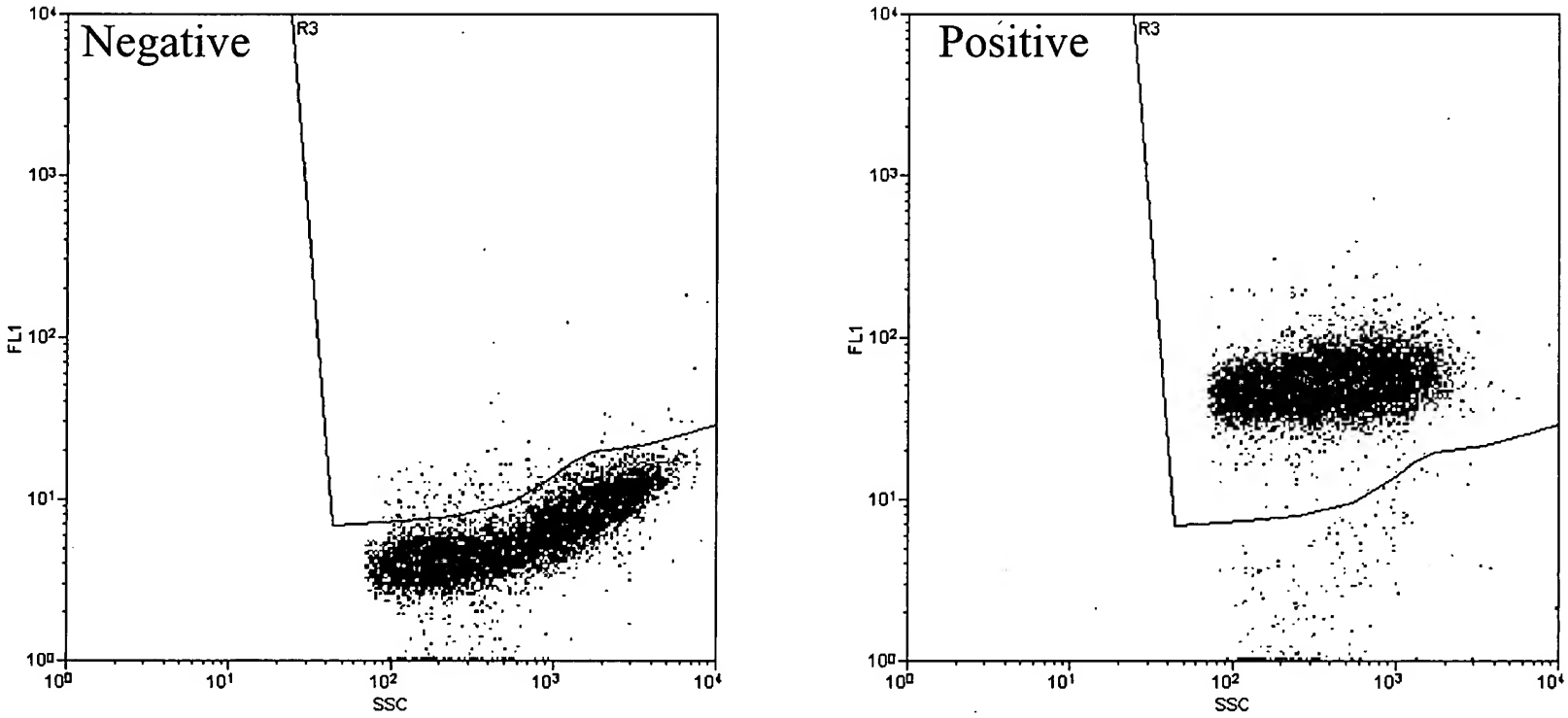


Fig. 30

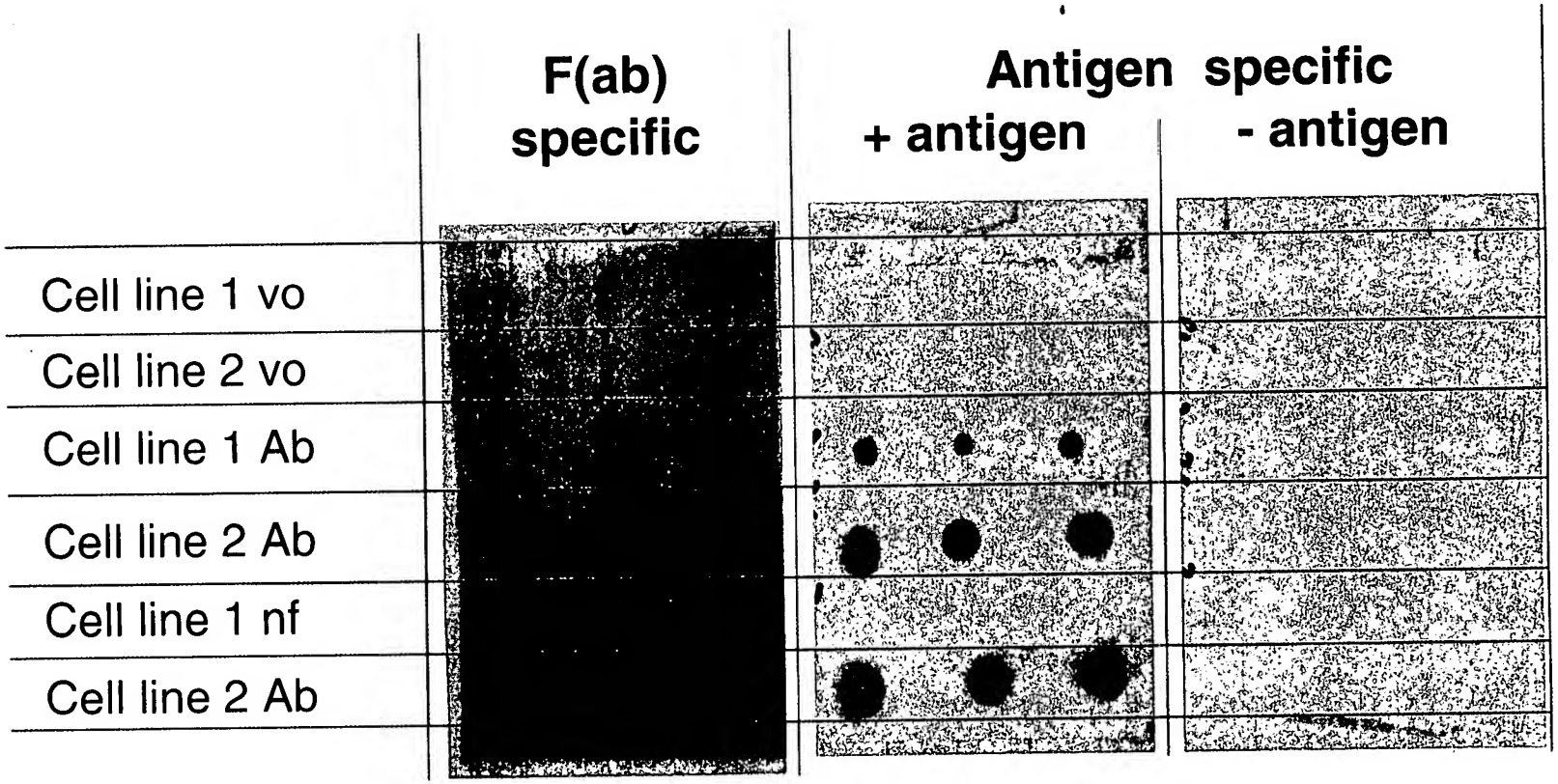

F(ab) specific
Antigen specific
+ antigen
- antigen
Cell line 1 vo
Cell line 2 vo
Cell line 1 Ab
Cell line 2 Ab
Cell line 1 nf
Cell line 2 Ab

Fig 31